CHAPTER 8
Sampling Design

Danaus plexippus
Monarch butterfly
Artist: D. Andrew Saunders

Design is critical to any sample-based monitoring study. The consequences of poor study design are many: lost time and money, reduced credibility, incorrect (or no) management decisions, and unnecessary resource deterioration, to name just a few. Take your time during this stage to design a study that will meet your management and sampling objectives in the most efficient manner.

Six basic decisions, which are discussed in detail in this chapter, must be made in designing monitoring studies based on sampling:

1. What is the population of interest?
2. What is an appropriate sampling unit?
3. What is an appropriate sampling-unit size and shape?
4. How should sampling units be positioned?
5. Should sampling units be permanent or temporary?
6. How many sampling units should be sampled?

These decisions must be made based on site-specific information and objectives. There is no "right" sampling-unit size and shape, just as there is no "right" number of sampling units. In most situations, these decisions can be made only through on-site assessment by pilot sampling.

Throughout this handbook we encourage you to initiate your monitoring study with a pilot study. This is essentially a trial run of your monitoring design. A pilot study accomplishes three critical things: 1) it provides estimates of the standard deviation needed to plug into sample size formulas to determine an adequate sample size to meet your sampling objective (Chapter 14); 2) it exposes problems at an early stage; and 3) it demonstrates whether a monitoring design is feasible. Based on the pilot study you perform, you may find that you cannot meet your objectives within the constraints of the time and money available. One solution to this dilemma is to change from sample-based monitoring to monitoring based on a qualitative technique or a complete census. Other solutions include choosing a different attribute to measure or changing your management and sampling objectives to reflect a less precise estimate (in the case of a target/threshold objective) or detection of a larger change (in the case of a change/trend objective).

The sampling-design issues discussed in this chapter pertain to monitoring studies in which all of the sampling units or individuals are available for measurement. In animal studies, individuals are often secretive and difficult to count. For these types of animals, most of the sampling-design issues discussed in this chapter are not applicable. Chapter 13 covers these situations.

WHAT IS THE POPULATION OF INTEREST?

As we learned in Chapter 7, the population consists of the complete set of units about which we want to make inferences. We are using "population" in the statistical, rather than the biological, sense. That both biologists and statistitians use the term "population" for different things creates ongoing confusion. To clarify the term, we describe four types of populations: biological populations, target populations, sampled populations, and statistical populations (Box 8.1).

A "biological population" is often difficult to define. A plant species that only occurs within a 100-hectare wetland with no other of this species found for over 100 km would likely be unanimously considered a biological population. A group of animals isolated on a single mountaintop would likely be considered unequivocally a population. Most plant and animal groupings, however, are less obviously isolated from others, creating a problem of identifying boundaries of the biological population. You will need to consider the biological population when assessing population rarity and risk (Chapter 3) and when developing ecological models that include immigration, emigration, and movement within and between biological populations (Chapter 14).

Management activities usually take place within some type of administrative boundary that does not respect the boundaries of the biological population. The portion of the biological population that you manage and are interested in we call the "target population." For example, if we

Box 8.1. *FOUR POPULATIONS*

A rare plant species grows in a 300 hectare wet meadow, isolated by about 40 km from the nearest of the 5 known occurrences of this species. Within the meadow, estimates of the number of individuals of this small perennial species range up to a million or more. A portion of the wet meadow (approximately 100 hectares) is managed by your office. This area was fenced 5 years ago to eliminate livestock grazing. The remainder is privately owned and lightly grazed; the landowner refuses to allow any monitoring on his land. You are limited to spending only 2 days per year monitoring the portion of the population managed by your office. You recognize that you cannot possibly sample the entire 100 hectares in a single day (the other day will be spent on data analysis and report-writing). Travel is difficult across the wet meadow, and you are concerned about disrupting a great heron rookery. You decide to establish a 100m × 100m monitoring site within the 100 hectares. Within this monitoring site, you will annually estimate density. After trials of several sizes and shapes of quadrats, you select a 25m × 0.5m quadrat for sampling, resulting in 800 potential quadrats that can be placed in the 100 × 100m area without overlap.

- *Biological Population: all plants within the 300 hectare wetland. (This is an easy example; most biological population boundaries are much more difficult to draw.)*
- *Target Population: all plants within the 100 hectares managed by your office.*
- *Sampled Population: all plants within the 100 × 100m monitoring site.*
- *Statistical Population: the 800 quadrats that may be potentially sampled.*

are interested in the success of a rare fish population as measured by the average length, our target population may be all the individuals of that species in a spring system of a preserve that has been set aside for that species' protection. Similarly, our target population might be all of the individuals of a rare plant species occurring within a particular wet meadow.

In sampling, the difference between the target population and the population you actually sample (the "sampled population") must be understood. When target populations are small and distributed in some uniform area such as all plants within a fenced pasture, we may be able to position sampling units throughout the entire target population. However, two factors usually lead to defining a new sampled population: 1) irregular target population boundaries, and 2) target populations that cover a very large geographic area.

When the target population is small, but has irregular boundaries, then we might fit some regular-shaped polygon such as a square or rectangle over the bulk of the population (as illustrated in Figure 8.1A). This newly defined area, often referred to as a **macroplot,** becomes our sampled population. The macroplot is usually permanently marked. The use of a macroplot facilitates the positioning of sampling units (see below) and ensures that the same area is sampled each year.

Macroplots are relatively large areas, with sampling units such as quadrats, lines or points randomly located within them.

We can make statistical inferences only to the boundaries of the sampled population (i.e., to the area within the macroplot), not to the entire target population. This approach works well for small target populations; a large population, however, would necessitate a very large macroplot, resulting in long distances between sampling units. The time necessary to travel to each sampling unit would make the design inefficient.

If the target population covers a very large geographic area, constraints of time and money, coupled with the tremendous variability usually encountered when sampling a very large popula-

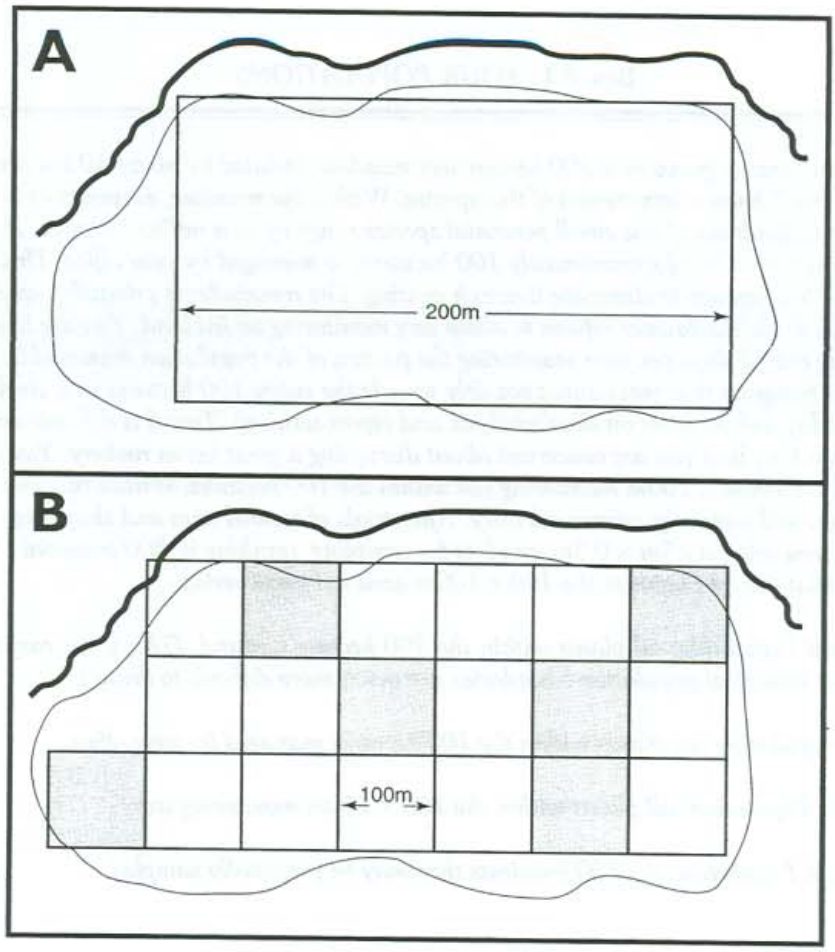

Figure 8.1. Positioning of macroplots (rectangles and squares) within irregularly shaped target populations (thin lines). The thick irregular line denotes a river. Figure 8.1.A. A single 200m × 75m macroplot is placed over the bulk of the target population. Inferences can be made only to the area within the macroplot (i.e., the macroplot is the sampled population). Figure 8.1.B. Target population covers a much larger area (note scale change). Six 100m × 100m macroplots are randomly placed within the target population. Inferences can be made to the entire target population (i.e., the sampled population is the same as the target population). Figure 8.1.C. A single square macroplot is placed in the target population. Inferences can be made only to the area within the macroplot (i.e., the macroplot is the sampled population). Figure 8.1.D. Subjective placement of a macroplot within a "representative" key area (dotted line).

tion, often require further restriction of the sampled population to a smaller geographic area. There are several ways this can be accomplished:

1. A sample of macroplots can be randomly positioned within the target population (Fig. 8.1B). If sampling takes place within each macroplot, then we have something called a two-stage sampling design, described in detail later in this chapter. Statistical inferences can be made to the entire target population, and the sampled population and the target population are the same.
2. A single macroplot can be subjectively positioned within the target population (Fig. 8.1C). The sampled population is the macroplot. No inferences to the target population are possible because there is no way of determining how "representative" this macroplot is of the target population.
3. A few macroplots can be subjectively positioned within the target population. Inferences can be made only to the area encompassed by the macroplots. In other words,

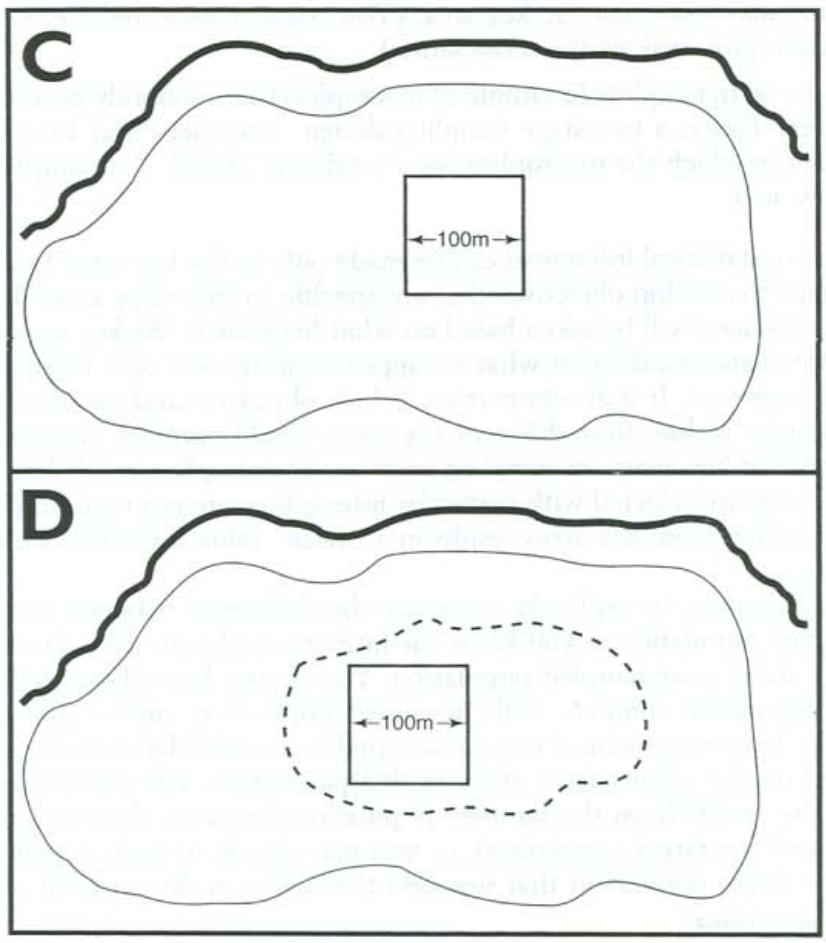

Figure 8.1. *(Continued)*

the sampled population is the area within the macroplots. The value of subjective positioning is that you can place the macroplots in the areas you consider most representative or critical.

When the target population area becomes very large and difficult to sample, we may select one or a few key areas in which we will conduct monitoring (Fig.8.1D). The key area concept is widely used, particularly in rangeland monitoring. Using this approach, key areas are selected (subjectively) that we hope reflect what is happening on a larger area. We may believe that the key area(s) are representative of a larger area (such as a pasture) or are critical or sensitive areas where we are most interested in detecting a problem.

Although we would like to make inferences from our sampling of key areas to the larger areas they are chosen to represent, this cannot be done statistically because the key areas were chosen subjectively. We could, of course, choose to sample the larger areas, but the constraints of time and money coupled with the tremendous variability usually encountered when sampling very large areas often make this impractical. The key area concept represents a compromise.

Careful definition of the sampled and target population remains critical. Remember the monitoring data only represents what is happening in the sampled population. Here are examples:

1. The key area is sampled with randomly placed quadrats. The key area is the sampled population.
2. One macroplot is subjectively positioned within the key area (Fig. 8.1D). You can only make inferences to the area inside the macroplot. Your sampled population is

the macroplot, not the key area. (You could reduce the size of the key area to the macroplot, making them the same.)

3. Several macroplots (a sample of macroplots) are randomly positioned within the key area. This is a two-stage sampling design. Inferences may be drawn about the key area in which the macroplots were randomly placed. The sampled population is the key area.

Because statistical inferences can be made only to the key areas that are actually sampled, it is important to develop objectives that are specific to these key areas. It is equally important to clarify that actions will be taken based on what happens in the key area, even when it cannot be demonstrated statistically that what is happening in the key area is happening in the area it was chosen to represent. It is also important to base objectives and management actions on each key area separately. Values from different key areas should never be averaged, because this gives the impression that key areas are sampling units used to sample a much larger area than is really the case. Key areas are selected with particular intent; they are not randomly selected sampling units. Averaging values from key areas results in a "mean" value for which we can have no measure of precision.

It is important to explicitly recognize the difference between your target population and your sampled population so you know the limitations of your data. You can only draw statistical inferences about your sampled population. You do not know how well the observations in the sampled population compare with the target population, unless you sample the entire target population. In management, it may be acceptable to make decisions for the entire target population based on the results from the sampled population. All stakeholders may have agreed to abide by the results from the sampled population (knowing there exists a risk that results may not represent the target population), or you may decide to collect qualitative or other ancillary data in the target population that supports the results in the sampled population. Consider the following questions:

- How limited are your monitoring resources?
- How difficult will it be to sample the entire target population?
- How comfortable will you (or the decision-maker) be in making management decisions for the entire target population based on the information gathered from a more limited sampled population?
- If the sampled area is located toward the middle of the population, will you miss changes that occur near the edge of the target population?

WHAT IS AN APPROPRIATE SAMPLING UNIT?

The type of sampling unit you select depends on the attribute you are measuring, which should be detailed in a specific management objective (see Chapter 14). Density, cover, frequency, biomass, and size of plant or animal populations are the attributes most commonly monitored. Attributes related to individual measures of performance such as height or number of flowers for plants and length and weight of animals are also often of interest (Box 8.2).

In many cases, simply determining the attribute you are going to measure determines the sampling unit. If you are going to measure density, frequency, or biomass, the sampling unit will be a quadrat. For cover, however, you have several choices. The sampling unit can be a line intercept, a point intercept, or a quadrat (Chapter 12 gives information to help you decide which of these to choose). If you are measuring something about individuals, the sampling unit is the individual (although, as we will see later, you will often incorporate quadrats). Most animals,

Box 8.2. *EXAMPLES OF SAMPLING UNITS*

- *Individual plants. Plants are the sampling units for attributes such as plant height, number of flowers per plant, or cover if the cover measurements are made on individual plants (e.g., tree stem diameters, bunchgrass basal area measurements).*

- *Individual animals. Animals are the sampling units for attributes such as height, length (e.g., snout-vent length in amphibians), condition (e.g., kidney fat index in ungulates), parasite loads, or reproductive rates (e.g., number of yearlings accompanying adult females).*

- *Plant parts. Fruits might be the sampling units if the attribute is the number of seeds per fruit or the percentage of fruits containing some seed herbivore. Or, you may be interested in estimating the number of flowers per inflorescence, in which case the inflorescence is the sampling unit.*

- *Quadrats (plots). Most estimates of plant density, frequency, or biomass require the use of quadrats, which represent the sampling units. Quadrats can also be the sampling units for measurements of vegetation cover if visual estimates of cover are made within quadrats. Most estimates of animal density, frequency, or biomass require the use of quadrats, sometimes called belt transects if greatly elongated, which represent the sampling units.*

- *Lines (transects). When cover is measured using the line-intercept method, the line is the sampling unit. Lines can also serve as sampling units when points (for cover) or quadrats (for cover, density, or frequency) are positioned along lines and the points or quadrats are not far enough apart to be themselves considered the sampling units (because they are not independent of one another). The line-intercept method is occasionally used to estimate animal populations based on the probability of transects intercepting animal tracks (Becker 1991).*

- *Points. When cover is measured with the point-intercept method and the points are randomly positioned, then the points are the sampling units. Points are sometimes used for sampling animals, mainly colonial ones that form large aggregations, such as corals.*

- *Point frames or point quadrats. When plant cover is measured using point frames or point quadrats and these frames or quadrats are randomly positioned, then the point frames or point quadrats are the sampling units. Point frames are not recommended because they are inefficient for measuring cover in most vegetation types (see Chapter 12).*

- *Distance (plotless) methods. There is a class of techniques to estimate density called distance or plotless techniques. The sampling unit with these techniques is usually the individual distance between a randomly selected point and the nearest plant or between a randomly selected plant and its nearest neighbor. Distance measures are inaccurate for most plant populations (see Chapter 12). Distance methods are also used in animal studies, but are different from those used in plant studies in that they attempt to overcome the problem of incomplete detectability of individual animals. These methods are discussed in Chapter 13.*

however, are too secretive or elusive to be directly counted; therefore, specialized sampling techniques (covered in Chapter 13) are needed to estimate most population parameters for animals.

Certain sampling designs incorporate sampling units at more than one level. These are called multistage sampling designs (Krebs 1998). The two-stage sampling design, discussed below, is one example. A random sample of primary sampling units is selected. Then, a subsample is taken from each of the primary sampling units. This subsample is made up of secondary sampling units (these are often called elements to differentiate between the two types of units).

The collection of sampling units from which you draw your sample is the statistical population. For example, a macroplot 20m × 50m will contain 4000 frequency quadrats 50cm × 50cm in size (quadrats do not overlap). The statistical population is the 4000 quadrats. If you were sampling with density quadrats 50cm × 25m in size, the statistical population is the total number of these that could fit into the 20m × 50m macroplot: 80 quadrats. These are finite statistical populations (see Chapter 7), unless so many potential quadrats exist within a large area that the number is essentially infinite. If you were sampling using line intercepts, the statistical population is all the potential line intercepts that could be placed within the 20m × 50m macroplot. Because lines have no width (theoretically, at least) an infinite number could be placed within the macroplot. The statistical population is thus infinite. This concept of infinite or finite populations has important implications for determining sample size and for analysis (see below and also Chapter 9).

WHAT IS AN APPROPRIATE SAMPLING UNIT SIZE AND SHAPE?

Considerations

The most efficient sampling unit size and shape depend on the type of attribute being measured and the morphology and spatial distribution of the species (or the object of your study such as nests, burrows, motorcycle tracks). The most efficient design is usually the one that yields the highest statistical precision (smallest standard error and narrowest confidence interval around the mean) for either a given area sampled or a given total amount of time or money available. Several factors must be considered:

Travel and Setup Time Versus Searching and Measuring Time

As the sampling unit increases in size, the time required to measure the unit increases. For estimating density in quadrats, for example, you must consider whether it is more important to minimize the number of sampling units or the total area (or proportion) of the population sampled. When sampling along transects, you must consider the time required to set up each transect, the travel time between them, and the time needed to measure each transect. Consider the size of the area you are sampling (is it a kilometer between each sampling unit?) and how difficult it is to get from one sampling unit position to another (are you sampling on a cliff face?). Also consider how hard it is to locate and measure the target species within each sampling unit. For large or conspicuous species such as large mammals, trees, or tall herbaceous plants that occur at low densities, having a large sample area or a long transect is not much of a problem because you can see all of the individuals, even from a distance. For small, obscure species that may be hidden under the vegetation canopy or under the leaf litter, you might have to search very carefully; in this case minimizing the total sample area or length may be critical.

Spatial Distribution of Individuals in the Population

Very few biological populations are randomly distributed in the area they occupy. If they were, different configurations of the same sampling unit size or length would perform similarly. Most populations, however, are aggregated or clumped in their distribution. For clumped distributions, sampling units that intersect some clumps of the species will reduce both the number of sampling units with zero counts and the number of sampling units with very high counts. This

decreases the variation among the quadrats and increases the precision of estimates. It is best if the sampling unit length (i.e., the length of the long side of the quadrat or the length of the transect) is longer than the mean distance between clumps.

As an example, consider the species *Primula wilcoxii*, which grows on the shaded side of terraces on a terraced slope in the foothills near Boise, Idaho. The terraces are approximately 1.5 meters apart. In this case, 1m × 1m quadrats to estimate density would be a very poor choice, because many of these would fall between terraces, resulting in many zero values. Some of the 1m × 1m quadrats, however, would fall right on the terraces, and very high counts of this species would be obtained for these quadrats. For this species at this terraced site, quadrats of 0.5m × 2.5m performed well.

Depending on the nature of your population, orientation of sampling units can be very important. For example, you want to orient rectangular quadrats to capture the variability within the quadrats rather than between the quadrats. This results in lower, among-quadrat variance and higher precision. Thus, if there is some gradient such as elevation or moisture to which the population responds differently, you want to make sure your rectangular quadrats follow that gradient to incorporate the variability within the quadrats. In the *Primula wilcoxii* example, the rectangular quadrats were most efficient when placed perpendicular to the terraces. If you were making counts of butterflies along transects,[1] you would orient the transects across changing habitats rather than run parallel to them. Similarly, if you were estimating cover using a line-intercept transect on a site that had a moisture gradient running from the east (wet edge) to the west (dry edge), you would orient your transects from east to west along the gradient.

Edge Effects

Edge effects are an important consideration for quadrat sampling units. The edge of a quadrat is its outer boundary. The more edge a quadrat has, the greater the difficulty in determining whether individuals near the edge are in or out of the quadrat. Rectangular quadrats have more edge per unit area than squares or circles. Although this is an important issue, Chapter 12 discusses ways to minimize the nonsampling error associated with edge bias when sampling plants (stationary animals would follow the same conventions). For most animals, determining whether an individual is in or out of the quadrat may be more difficult because they are moving and because you usually cannot necessarily measure the distance to them. This is, for example, an important issue when counting birds, some of which are often entering and leaving a study plot while a count is being made. Chapter 13 discusses these issues. You must be consistent in applying whichever boundary convention you choose and to make sure, through training and documentation, that others involved in the monitoring during the first and all subsequent years use the same convention.

Abundance of Target Population

If the density is relatively high, you will want to use smaller quadrats because you do not want to be counting hundreds to thousands of occurences in each quadrat. Conversely, if density is relatively low, you will want to use larger quadrats to avoid sampling many quadrats with no individuals in them.

Ease in Sampling

The considerations here are the difficulties in searching the entire sampling unit and keeping track of what portions have already been searched. With large quadrats for measuring density, for example, long, narrow rectangles are easier to search because you can start at one end and keep track of counts at intervals along the quadrat. With large, square quadrats, you will probably have to subdivide the quadrat area to ensure that you do not double-count.

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[1] *These types of transects in which counts are made are actually very long, narrow quadrats, compared to true transects which have no width (theoretically dimensionless).*

Disturbance Effects

If the sampling unit size/shape is so large that you have to stand in the sampling unit to search through it, you risk impacting the population through your sampling. This is particularly important when sampling permanent sampling units, because the changes you observe over time may simply be the result of your impacts to the sampling units and not reflect the true situation in the sampled population as a whole. It is also a problem when using temporary sampling units, however, especially if you impact areas of the sampling unit before you have searched them.

Computer-Simulated Comparisons of Sampling Designs

The importance of selecting an efficient sampling unit configuration is often ignored when developing a monitoring study. Sampling units of different configurations perform differently, and the efficiencies to be realized from using an appropriate configuration can be substantial. We will use a particular type of sampling unit, quadrats for estimating density, to explore this issue further using computer simulation.

As stated earlier, rectangular quadrats perform better than square or circular quadrats when sampling clumped populations, but two unanswered questions remain: 1) What are the actual trade-offs of changing quadrat size and shape on the number of quadrats to sample or on the total area sampled? 2) As you make quadrats larger, you will presumably have to sample fewer of them—but how many fewer? You can investigate these questions in the field, but you are somewhat limited in the number of different sizes and shapes you can try, and there are potential negative impacts from repeated sampling across the entire area.

Salzer (unpublished data) evaluated these sampling design decisions using computer-simulated sampling. Two populations, each with 4000 plants, were created on the computer. Plants in both populations exhibited a clumped distribution pattern, although they differed in the degree of clumping. One of the populations had plants that were distributed along a gradient. Random samples of these virtual populations were drawn by computer, using density quadrats of different sizes and shapes.

Consider the population of 4000 plants depicted in Figure 8.2. This population was termed the "clumped-gradient population" because the plants were both clumped and distributed along a gradient (note that this gradient follows the x-axis: there are more clumps near the left side of the macroplot than there are near the right side). This population was subjected to 30 different sampling designs that differed in the width and length of the quadrats. The following quadrat widths were used: 0.25m, 0.5m, 1.0m, 2.0m, and 4.0m. The following quadrat lengths were used: 1m, 2m, 5m, 10m, 25m, and 50m. Every combination of quadrat width and quadrat length was used to sample the population (i.e., 0.25m × 1m, 0.25m × 2m . . . 4m × 25m, 4m × 50m). Sampling was conducted so that the long side of the quadrat was oriented so the quadrat included as much of the changing gradient as possible (i.e., the long side was oriented parallel to the x-axis of the population as depicted in Figure 8.2).

For each of the 30 sampling designs, the entire population was sampled (i.e., all the quadrats that fit in the population, without overlapping), so that true, parametric values for the mean density and standard deviation could be calculated for every design. This is desirable for comparing various sampling designs, but is nearly impossible to achieve in a field setting. The true parametric values were entered into a sample size formula to determine how many quadrats would need to be sampled to attain the desired precision. The precision level selected was an estimated mean density with a 95% confidence interval that was no wider than ±30% of the mean value. This brought performance of each sampling design into a common currency—the number of quadrats to sample—so that they could be compared with one another. By knowing the size and number of quadrats being used, the proportion of the population sampled was also calculated.

Figure 8.3 depicts the interaction between quadrat width, quadrat length, number of quadrats, and proportion of the population sampled. A typical quadrat configuration used in

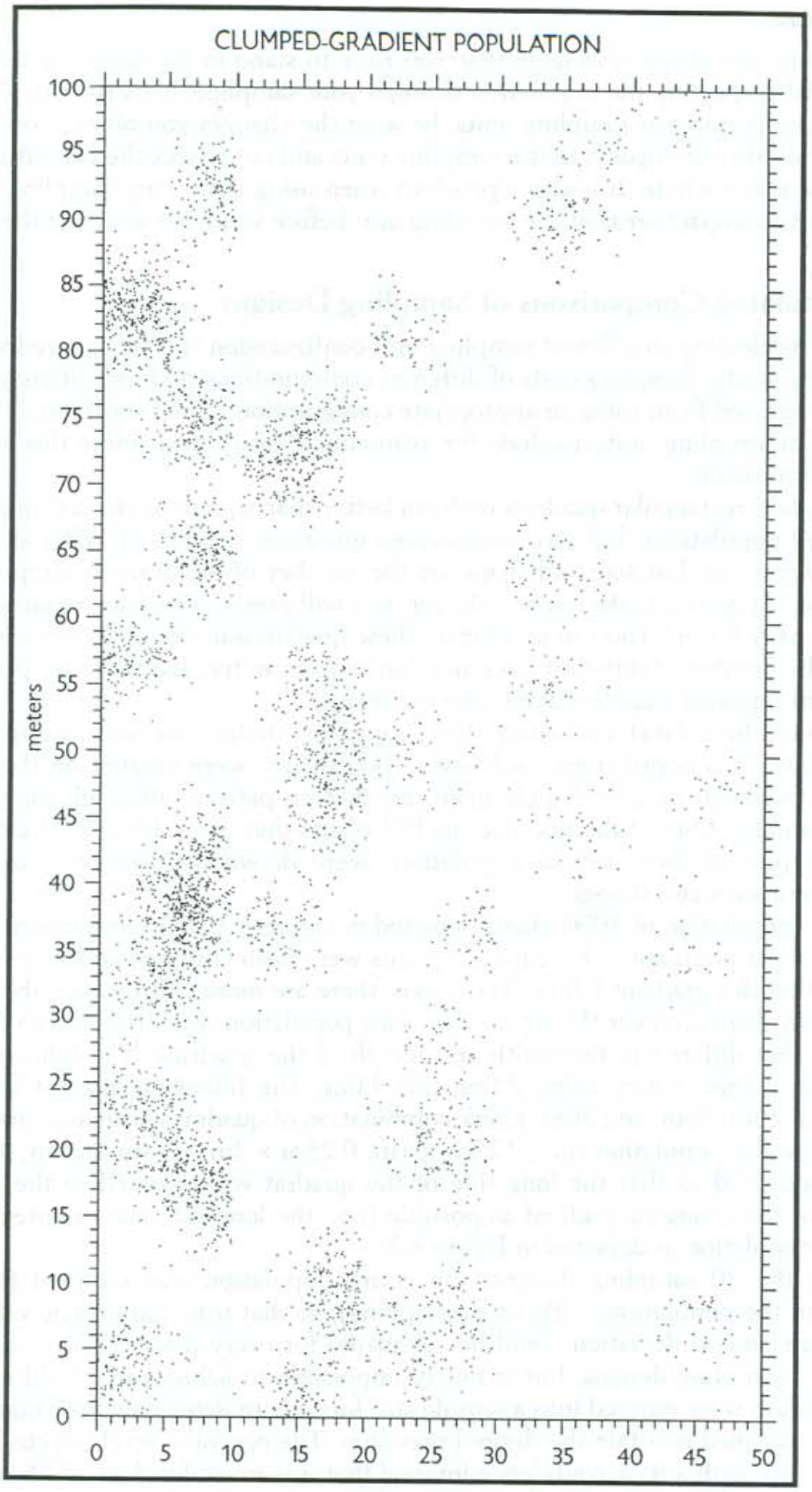

Figure 8.2. The "clumped-gradient population." A population of 4,000 plants aggregated into clumps and responding to a gradient that runs from left to right (along the x-axis). Note the much greater number of clumps near the left side of the population.

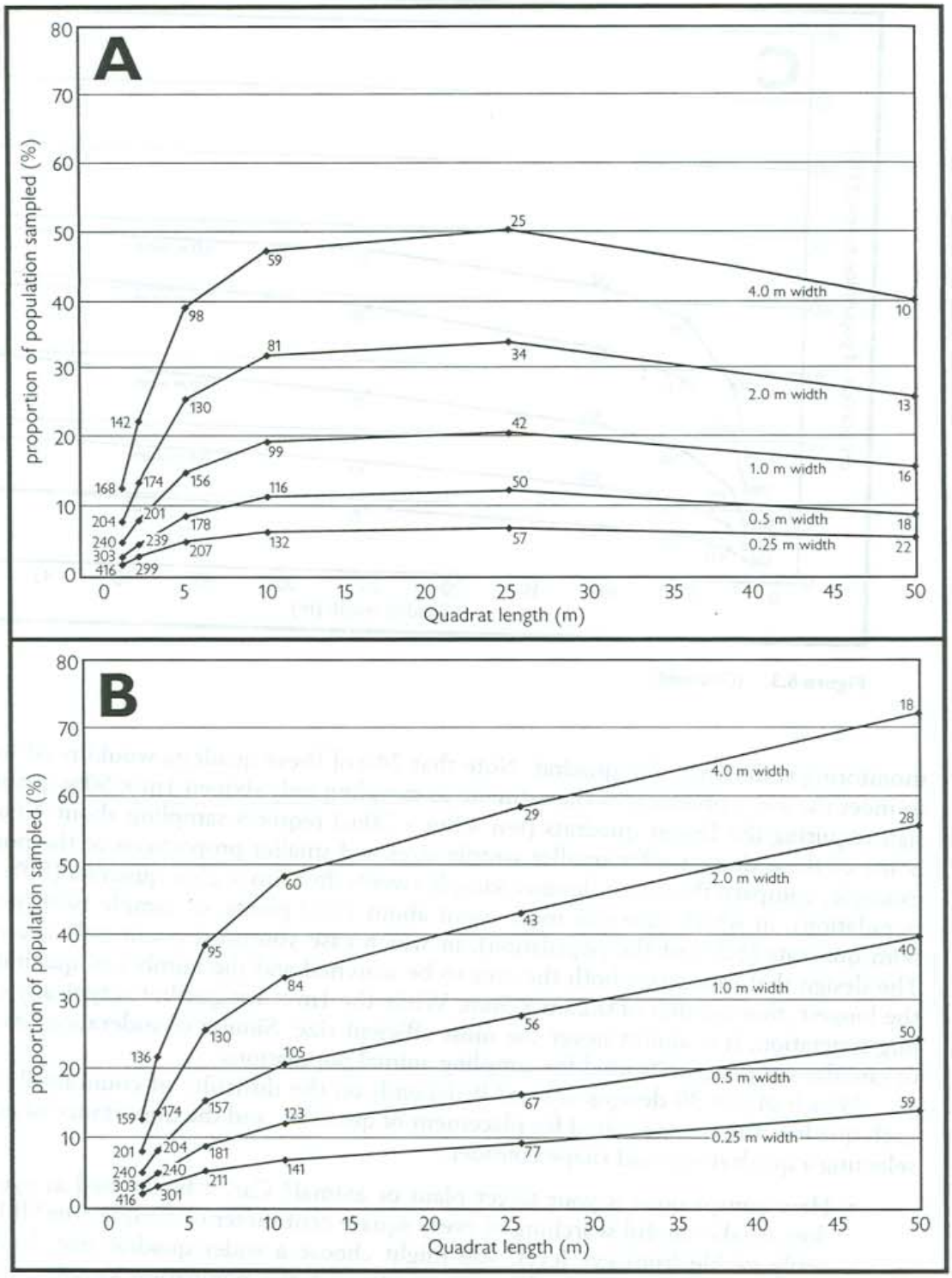

Figure 8.3. Comparison of 30 sampling designs to sample density. Designs used quadrats of different widths (0.25m, 0.5m, 1.0m, 2.0m and 4.0m) and lengths (1m, 2m, 5m, 10m, 25m, 50m) for a total of 30 different quadrat configurations. All designs achieved the same level of precision. Numbers next to data points are the number of quadrats that must be sampled to meet the desired level of precision in the estimate of mean density using that particular quadrat size and shape. Figure 8.3A shows the results when quadrats are oriented along the gradient shown in the population of Figure 8.2 (i.e., the long edge of the quadrat along the x-axis of the population, including as much of the gradient variability as possible). Figure 8.3B shows the results when quadrats are oriented perpendicular to the gradient (i.e., the long edge of the quadrat along the y-axis of the population) shown in Figure 8.2. Figure 8.3C shows the results from sampling a similar population of 4000 plants that lacks a gradient but has much denser clumping (i.e., more unoccupied space between clumps). This densely clumped population is shown in Figure 8.5.

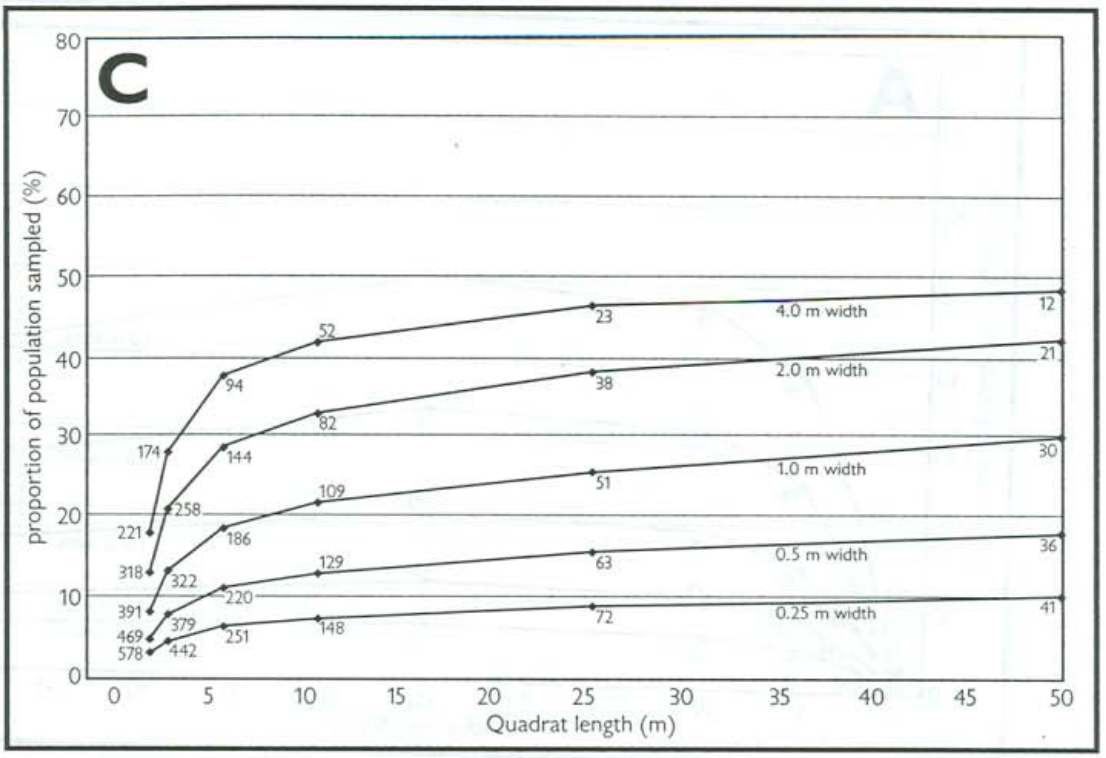

Figure 8.3. *(Continued)*

monitoring is the 1m × 1m quadrat. Note that 240 of these quadrats would need to be sampled to meet the same precision of the estimate as sampling only sixteen 1m × 50m quadrats. The design requiring the fewest quadrats (ten 4.0m × 50m) requires sampling about 40% of the area. Some of these designs offer smaller sample sizes and smaller proportions of the population. For example, compare these two designs: sample twenty-five 4m × 25m quadrats (50% of the entire population), in which case you must count about 2000 plants, or sample twenty-two 0.25m × 50m quadrats (5.5% of the population), in which case you must count only about 220 plants. The design that minimizes both the area to be searched and the number of quadrats to locate is the longest, thin quadrat of 0.25m × 50m. While the 1m × 1m quadrat is typically used in sampling vegetation, it is almost never the most efficient size. Similar considerations exist for square or circular sampling units used for sampling animal populations.

Which of the 30 designs is best? It depends on the difficulty of counting the plant within each quadrat, the time required for placement of quadrats, and the importance of edge effects. In selecting a quadrat size and shape consider:

- How conspicuous is your target plant or animal? Can it be spotted at eye-level or does it take careful searching of every square centimeter of sample area? If large and easily visible from eye level, you might choose a wider quadrat size, leading to a smaller sample size. The larger proportion of the population sampled might not carry much of a penalty (cost) if the portions of the quadrats between clumps can be searched rapidly.
- How quickly can you locate sampling units? If travel between sampling units is difficult because of topography or dense vegetation, sampling fewer larger quadrats would probably save time.
- How big a problem is edge effect? Are plants single-stemmed with small diameter stems clearly arising from a rooted point so that boundary decisions are relatively

rare and quickly made when they do occur or are the target plants bunch grasses with a wide basal area and amorphous shapes, requiring many difficult and time-consuming boundary decisions? Are the animals relatively slow-moving, or is determining whether they are in or out of the sampling unit difficult because they are moving quickly?

If plants are small and inconspicuous with distinct, single-rooted stems, look for a design that has both a small sample size and samples a small proportion of the population. The twenty-two 0.25m × 50m quadrats would be a good choice in this case. Realize, however, that even if minimizing the sample area is critical, you will not want to sample 416 of the 0.25m × 1m quadrats (2.1% of the population area).

Results for the same clumped-gradient population with quadrat orientation reversed (i.e., with the long side parallel to the y-axis) are shown in Figure 8.3B. Rather than looking at the individual sample sizes, concentrate on just the relative proportion of the population that must be sampled. With this quadrat orientation, quadrats located near the left of the macroplot will have high numbers of plants, while quadrats located near the right of the macroplot will have low numbers. This pattern of high and low quadrat counts is undesirable, producing a high standard deviation and wide confidence intervals. With the 4m × 50m quadrat, you need to sample over 70% of the population. You would be better off counting all of the plants in the macroplot (conducting a complete census) than using this quadrat size. Clearly it is better to use a narrower quadrat that is oriented in the opposite direction.

Results from a population of 4000 plants that are more tightly clumped with the clumped centers randomly distributed (without a gradient) are shown in Figure 8.3C. (You can see this population in Figure 8.5). Because of the tighter clumping of plants in the dense-clumped population, sample sizes are even greater for small and square or short and wide quadrats than they were for the clumped-gradient population. This is because quadrats with plants tend to have higher counts and there are more quadrats with zero plants, a situation that increases the standard deviation. It would take, for example, 578 of the 1m × 0.25m quadrats to achieve the desired level of precision in the dense-clumped population as compared with 416 in the clumped-gradient population. With increasing clumping, the advantages of long, narrow quadrats also increase. Conversely, if plants are randomly distributed, quadrat shape has no influence on the number of quadrats to sample. This, however, is seldom the case in nature.

Even though the narrower quadrat sizes perform better statistically, there are practical limitations that must be considered. For example, when sampling the virtual dense-clumped population by computer using different shapes of quadrats with an area of $1m^2$, a 2cm-wide × 50m-long quadrat performed better (n = 98 quadrats) than a 1m × 1m quadrat (n = 394), but the 2cm width would be a ridiculous shape to try to use in the field, because of the tremendous amount of "noise" introduced by edge effect.

In many monitoring situations, especially for herbaceous plants or small, slow-moving animals, a 0.25m or 0.5m quadrat width works well for estimating density. (This width would probably be inappropriate, however, for large or sparsely distributed plants, or for large or fast-moving animals.) Either is a convenient width to search in. Widths larger than 1m or 2m are difficult to search because it is hard to see individuals at the far edge (unless all the individuals are fairly large and there is minimal associated vegetation to obscure your line of sight). The quadrat length should be determined by the size of the area that you are working in and the spatial distribution of the species you are counting. You want to avoid getting many sampling units with zeros, so you want your quadrats to be long enough to incorporate several clumps. You also do not want your quadrats so long that you have to count thousands of individuals—the time involved and the potential measurement error associated with counting that many individuals would be too great. Box 8.3 gives a procedure for comparing the efficiency of different density quadrat sizes and shapes through pilot sampling.

Box 8.3. *A PROCEDURE TO COMPARE THE EFFICIENCY OF DIFFERENT QUADRAT SIZES AND SHAPES USING PILOT SAMPLING*

Select several good candidates of quadrat dimensions that are multiples of the two dimensions of the area you want to sample. For example, in a 50m × 100m macroplot where you want to orient quadrats with the long side along the 50m side of the macroplot, you might select 5m, 10m, 25m and 50m (all factors of 50m) and widths of 0.25cm and 0.50cm. Randomly locate some initial number (e.g., 10) of 0.5m × 50m quadrats in the population of interest. Position the quadrats according to the design you plan to use (this will allow you to use the data from this initial test as part of your actual sample). Attach one end of a 50m tape to a pin or stake, pull it tight and treat one edge of the tape as the center of your quadrat. Count all plants that are within 0.25cm of either side of the tape edge (total width = 0.5m) and record separately, by side, on a field data sheet (Figure 8-B). You should also subdivide the long dimension of the quadrat and record plant counts separately within each segment (e.g., every meter) along your tape. This enables you to look at the performance of quadrats of different lengths.

You can save space by recording the segment number only if you have actual plant counts for that segment. For example, you have laid out your tape and started searching along both sides of the tape. You find your first plants (three of them on the left side and two of them on the right side in the third segment of the tape (between 2m and 3m along the tape). The next plants (two of them on the left side, none on the right) are found in the seventh segment (between 6m and 7m along the tape). The entries on the field data sheet would look like Figure 8-B.

Continue this counting and recording procedure until all your preliminary quadrats have been sampled. Now you can use a hand calculator to calculate means and standard deviations for different size and shape quadrats. To compare quadrats of different sizes you should calculate the coefficient of variation (CV) for each quadrat size. The CV is calculated as follows:

$$CV = s/\bar{x}$$

Where: $\bar{x}$ = The sample mean
s = The sample standard deviation

Unlike the standard deviation, which has a magnitude dependent on the magnitude of the data, the coefficient of variation is a relative measure of variability. Thus, coefficients of variation from different sampling designs can be compared. The smaller the coefficient of variation the better. If two designs have similar coefficients of variation, choose the design that will be easiest to implement.

If, after evaluating the performance of different quadrat sizes, you select a size and shape that was some subcomponent of the larger quadrat sampled, you can still use the data as part of your first year's set of data. To do this you should randomly select the subcomponent from each of your pilot quadrats. Using the previous example, if you elected to use a 0.25m × 50m quadrat, you could randomly select one half of each of the 0.5m × 50m quadrats that you sampled as part of your pilot effort.

PLOT #	SEGMENT #	PLANT COUNTS		
		Left	*Right*	*Total*
1	3	3	2	5
1	7	2	0	2

Figure 8-B. Examples of entries on a field data sheet when plants are found in the third and seventh segments of plot number 1.

Other Sampling Units

Your prime design objective when selecting a sampling-unit size and shape is to try to reduce the variability between sampling units while maintaining a size and shape that is practical in the field. Many of the design principles described for density quadrats are applicable to other types of sampling units. Transects should be long enough to intersect clumps of the target species and should be oriented to include as much of the gradient variation as possible. Plots for visually estimating cover or measuring biomass are typically quite small and often square or rectangular, because it is difficult to estimate cover, to clip vegetation, or to estimate biomass in large or long plots. These small quadrats can be arranged, however, along a transect, with the transect, not the quadrats, treated as the sampling unit. This design is really a two-stage sampling design, with the transects serving as the primary sampling units and the quadrats serving as secondary sampling units. We treat this in more detail below.

Chapter 12 describes sampling-unit design considerations for most of the typical methods of measuring plants: density, cover measured by point intercept, line intercept and quadrat estimation, biomass measurements, and frequency measurement. Chapter 13 describes special considerations in sampling-unit design for animal studies.

Determining Sampling-Unit Size and Shape in Real Populations

The best way to determine the appropriate sampling-unit size and shape is to approach every new sampling situation without a preconceived idea of the configuration you will use. Sampling-unit size and shape should be determined during pilot sampling. If possible, wander around the population area and study the spatial distribution of the species you will be sampling (for plants, use pin flags or flagging to improve the visibility of clumps). Attempt to answer the following questions: 1) At what scale(s) can you detect clumping? 2) How large are the clumps, and what are the distances between clumps? 3) How long will sampling units need to be to avoid having many sampling units containing none of the species in them? 4) How narrow will density quadrats need to be to avoid counting hundreds or thousands of the species whenever the quadrat intersects a dense clump? 5) How wide an area can be efficiently searched from one edge of a quadrat? 6) How big a problem will edge effect be?

HOW SHOULD SAMPLING UNITS BE POSITIONED IN THE POPULATION?

There are three requirements that must be met by a monitoring study with respect to positioning sampling units in the population to be sampled: 1) some type of random, unbiased sampling method must be employed; 2) the sampling units must be positioned to achieve good interspersion of sampling units throughout the population; and 3) the sampling units must be independent of each other. Before discussing different methods of random sampling, we will discuss these three characteristics in more detail.

1. Random (unbiased) sampling. Critical to a valid monitoring study design is that the sample has been drawn randomly from the population of interest. Several methods of random sampling can be used, many of which are discussed below. The important point is that all the statistical-analysis techniques available to us are based on knowing the probability of selecting a particular sampling unit. If some type of random selection of sampling units is not incorporated into your study design, you cannot determine the probability of selection, and you cannot make statistical inferences about your population. Preferential sampling, the practice of subjectively selecting sampling units, should be avoided at all costs.

2. Interspersion. One of the most important considerations in sampling is good interspersion of sampling units throughout the area to be sampled (the target population). Although Hurlbert (1984) uses the term "interspersion" to apply to the distribution of experimental units in manipulative experiments, the term can also be applied to sampling units in observational studies. The basic goal is to have sampling units well interspersed throughout the area of the target population. For this reason, the practice of placing all the sampling units, whether quadrats or points, along a single or even a few transects must be avoided. This is true even if the single transect or few transects are randomly located.
3. Independence. Independence means the sampling units are spaced far enough apart so that measurements are not spatially correlated. For example, if quadrats are not spatially correlated, high mortality in Quadrat A does not necessarily mean there will be high mortality in Quadrat B, at least not because of its proximity to Quadrat A. If your design has quadrats located closely along a transect, each quadrat is in close proximity to two others, and changes in each quadrat will probably be correlated with two others (or more). In simple random sampling, there will always be some quadrats located close together simply by chance. The difference is that this correlation only affects some of the quadrats, and the degree of correlation fluctuates randomly with the spatial location of the randomly placed quadrats.

We discuss eight types of random sampling: simple random sampling, stratified random sampling, systematic sampling, restricted random sampling, cluster sampling, two-stage sampling, double sampling, and taking a random sample of individuals. These are summarized in Table 8.1 and are described in more detail below.

Table 8.1. Summary of Random Sampling Types

SAMPLING TYPE	RECOMMENDED USES	ADVANTAGES	DISADVANTAGES
Simple random sampling	Useful in relatively small geographic areas with homogeneous habitat, when the number of sampling units is not likely to be large.	The formulas necessary to analyze data are the simplest of all sampling types.	By chance, some areas within the target population may be left unsampled. The travel time is considerable when the sampling area and/or sample size is large. Restricted random sampling and systematic random sampling outperform simple random sampling when populations have a clumped distribution.
Stratified random sampling	Useful when the attribute of interest responds very differently to some clearly defined habitat features. Since it involves taking a simple random sample within each stratum, each stratum should consist of a relatively small geographic area with homogenous habitat, and the number of sampling units in each stratum should not be too large.	Results in more efficient population estimates than simple random sampling when the attribute measured varies with clearly defined habitat features.	The mathematic formulas required for analysis are more complex than those used for simple random sampling. When the geographic area within any stratum is large and/or the number of sampling units is likely to be large, then one of the other types of sampling listed below will be more efficient. By chance, some areas within each stratum may be left unsampled.
Systematic sampling	Useful for any sampling situation, as long as the first sampling unit is selected randomly and the sampling units are far enough apart to be considered independent. Can also be used as part of cluster and two-stage sampling designs.	When the conditions given in the cell to the left are met, this is the best type of sampling design to use. There is better interpersion of sampling units than with simple random sampling. The data can be gathered much more efficiently than with simple random sampling and still be analyzed using the formulas for simple random sampling.	In the uncommon event that the number of possible samples is limited to fewer than about 25–30 (see text), systematic sampling may lead to questionable results; in this situation you should use restricted random sampling.

(Continued)

Table 8.1. Summary of Random Sampling Types (*Continued*)

SAMPLING TYPE	RECOMMENDED USES	ADVANTAGES	DISADVANTAGES
Restricted random sampling	Although more useful than simple random sampling in most situations, restricted random sampling should be used only when the number of potential samples is fewer than 25–30. Otherwise, systematic sampling is the better choice.	Like systematic sampling, restricted random sampling results in better interspersion of sampling units than with simple random sampling. If the number of potential samples is less than 25–30, restricted random sampling is better than systematic sampling. The data can be analyzed using the formulas for simple random sampling.	The design is not as efficient as systematic sampling when the number of potential samples is greater than 25–30.
Cluster sampling	Cluster sampling is used to select a sample when it is difficult or impossible to take a random sample of the individual elements of interest. A cluster of elements is identified, and a random sample (usually using systematic sampling) is taken of the clusters. Every element within each cluster is then measured. In monitoring, cluster sampling is most often used to estimate something about individuals (e.g., mean height, number of flowers/plant). In this situation, quadrats are the clusters.	It is often less costly to sample a collection of elements in a cluster than to sample an equal number of elements selected at random from the population. Except in rare situations, it is not practical to take a random sample of individuals. Instead, the attribute of interest is measured on every individual in a sample of quadrats (which function as the clusters).	All the elements within each cluster must be measured. If the clusters contain large numbers of the element of interest, two-stage sampling is more efficient. Other disadvantages include the difficulty in determining how many clusters should be sampled versus how large each cluster should be, the more complex calculations required for analysis, and the fact that most statistical software packages do not include these calculations.
Two-stage sampling	Similar to cluster sampling in identifying groups of elements (such as plants) and taking a random sample (usually using systematic sampling) of these groups. In two-stage sampling, however, a second sample of elements is taken within each group. Like cluster sampling, the main use of two-stage sampling is to estimate some value associated with individuals.	Same advantages as cluster sampling. The two types are the only efficient means of estimating some attribute associated with individuals. When the number of individuals in each group (quadrat) is large, two-stage sampling is more efficient than cluster sampling.	There are standard deviations associated with both stages of sampling (unlike cluster sampling, which has no standard deviation associated with the values measured at the second stage). This results in more complicated formulas in arriving at estimates of values and standard errors (although the standard deviation of the secondary sample can be ignored as long as the finite-population correction factor is not applied to the standard error of the primary sample).
Double sampling	Useful when the variable of interest (e.g., actual measurements of biomass) is difficult to measure, but is correlated with an auxiliary variable (e.g., ocular estimates of biomass) that is more easily measurable. The second variable is measured in a large number of sampling units, while the first variable is measured in only a subset of the sampling units. The samples are often taken using systematic sampling.	If the auxiliary variable is relatively quick to be measured and is highly correlated with the variable of interest, double sampling is much more efficient in estimating a variable that is difficult to measure than directly measuring the variable.	The formulas for data analysis and sample-size determination are much more complicated than for simple random sampling, and most statistical software programs do not include the necessary calculations.
Taking a random sample of individuals	This can only be accomplished in rare situations. When the objective is to measure something on individual plants, it is best to use either cluster or two-stage sampling. See text for further information.	In those few situations where it is possible to take a random sample of individuals, the calculations necessary for analysis are simpler than those for either cluster or two-stage sampling.	It is not practical to take a simple random sample of individuals in most monitoring situations.

Simple Random Sampling

A simple random sample is one that meets the following two criteria: 1) each combination of a specified number of sampling units has the same probability of being selected; and 2) the selection of any one sampling unit is in no way linked to the selection of any other (McCall 1982).

One method for selecting random sampling units in a simple-random-sampling design is the **simple random-coordinate method**. While this is probably the most commonly used method, it has serious problems for many sampling units. As shown in Figure 8.4, random coordinates are selected for each of two axes. The point at which these intersect specifies the location of a sampling unit. Coordinates that fall out of the target population boundaries are rejected. This method will work for small sampling units such as plots used to measure frequency,[2] but it will not perform well when the sampling units are lines or long rectangles or when sampling units are points of the center of a large circular sampling unit (e.g., bird counts often sample a circular area with a radius of up to 100m extending from a given sampling point, which often results in a large part of the sampled area falling outside a study site). Two problems with the coordinate method are difficult to overcome:

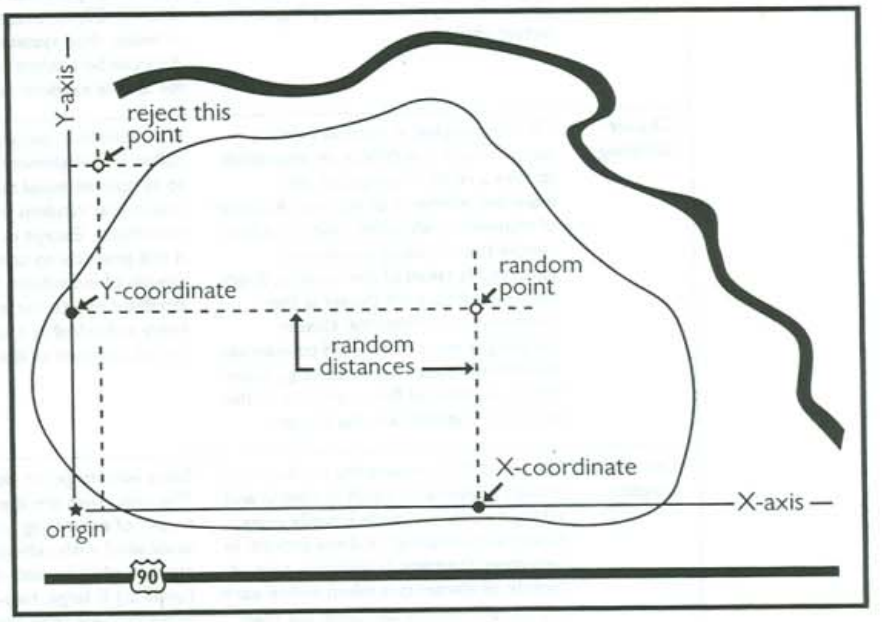

Figure 8.4. Locating points using the simple random coordinate method (adapted from Chambers and Brown 1983). Although this method will work to position points or square quadrats, the grid-cell method is much better for locating long, narrow quadrats or lines.

1. No unbiased method exists to deal with randomly located points that send a portion of the sampling unit out of the target population (a common occurrence with large or long sampling units). If you reject such points, your sample will be biased toward the center of the population (i.e., you will be less likely to sample the edges of the population). If you "reflect" the line or quadrat from the population edge back into the population, you bias your sampling toward the edges of the population.
2. This technique introduces the probability of overlapping sampling units. This is, for example, a major problem with bird surveys, in which some birds can be detected up to 100m away, necessitating that sampling points be separated by twice that distance. For quadrats (either rectangular ones, or circular ones located by their center point) overlap is highly undesirable, because we will not be able to use the finite-population correction factor discussed later in this chapter. For transect line intercepts, you could address overlap by selecting random compass orientations from each randomly located point; lines represent an infinite population regardless of their orientation and so we never use the finite population correction factor. This

[2] *Although such a random selection procedure is justified for sampling with point intercepts, frequency quadrats and biomass and cover estimation quadrats, the time required to position 100 to 200 or more of these small sampling units makes this procedure impractical. Instead, some type of systematic approach is usually used.*

approach, however, eliminates the possibility of orienting lines consistently along the gradient.[3]

A better method for locating random sampling units is the **grid-cell method**. The grid-cell method eliminates the problems associated with the random coordinate method and is one of the most efficient and convenient methods of randomly positioning quadrats. The sampled population area is overlaid with a conceptual grid (there is no need to actually lay out tapes and strings to achieve this), where the grid-cell size is equivalent to the size of each sampling unit. Consider the dense, clumped population example introduced earlier. We have overlaid a grid of 4m × 10m quadrats on this population (Fig. 8.5). If we want to sample ten 4m × 10m quadrats from this population, we would first divide the population into 125 different 4m × 10m cells, as shown on Figure 8.5. Since we are sampling without replacement, 125 possible quadrat positions (5 along the x-axis times 25 along the y-axis) are possible, none of which overlap. Once one is sampled, it will not be sampled again (at least not during the same sampling period). More information on implementing the grid-cell method in the field is given in Chapter 11.

As its name suggests, simple random sampling is the simplest kind of random sampling, and the formulas used to calculate means and standard errors are easier than with many of the more complex types of designs discussed below. But unless you are planning to use permanent quadrats to detect change, simple random sampling should only be used in relatively small geographic areas where a degree of homogeneity is known to exist. If the sampling area is large and/or the sample size is relatively large, as it often is for frequency or point-intercept simple random sampling, the time spent in locating quadrats or points and traveling between locations can be considerable.

Another problem with simple random sampling is that, simply by chance, some areas may be left unsampled. Figure 8.6 shows a simple random sample of a hundred 1m × 1m quadrats positioned within a 50m × 100m macroplot. By chance, some large portions of the macroplot did not receive any sampling units. This can be especially problematic in populations that are clumped. Computer-simulated sampling (Salzer, unpublished data) suggests that both restricted random sampling and systematic sampling designs (described below) result in more precise estimates than simple random sampling when sampling clumped distributions (the most common situation in biologic populations).

Stratified Random Sampling

Stratified random sampling involves dividing the population into two or more subgroups (strata) before sampling. Strata are generally delineated in such a manner that the sampling units within the same stratum are very similar, while the units between strata are very different. Simple random samples are taken within each stratum.

Strata should be defined based on the response (of the attribute that you are estimating) to habitat characteristics that are unlikely to change over time. Examples of characteristics that might be used to delineate strata are soil type, aspect, major vegetation type (e.g., forest or grassland), and soil moisture. You should avoid defining strata based on characteristics related to the attribute you are estimating, since this is likely to change with time, leaving you stuck with strata that are no longer meaningful. For example, if you are interested in estimating the density of species X, and you note that the east half of the target population is much more densely populated than the west half, avoid basing your strata on this fact alone. If there is an obvious habitat feature responsible for this difference such as aspect, then base your strata on this habitat feature. If there is no obvious reason for the difference, you are probably better off using a simple-

[3] *You should orient sampling units to include as much of the gradient variation as possible within the sampling unit. This maximizes variability included within the sampling unit and minimizes the variability between them, and can dramatically increase the efficiency of the sampling design. See the computer-simulated sampling design example above.*

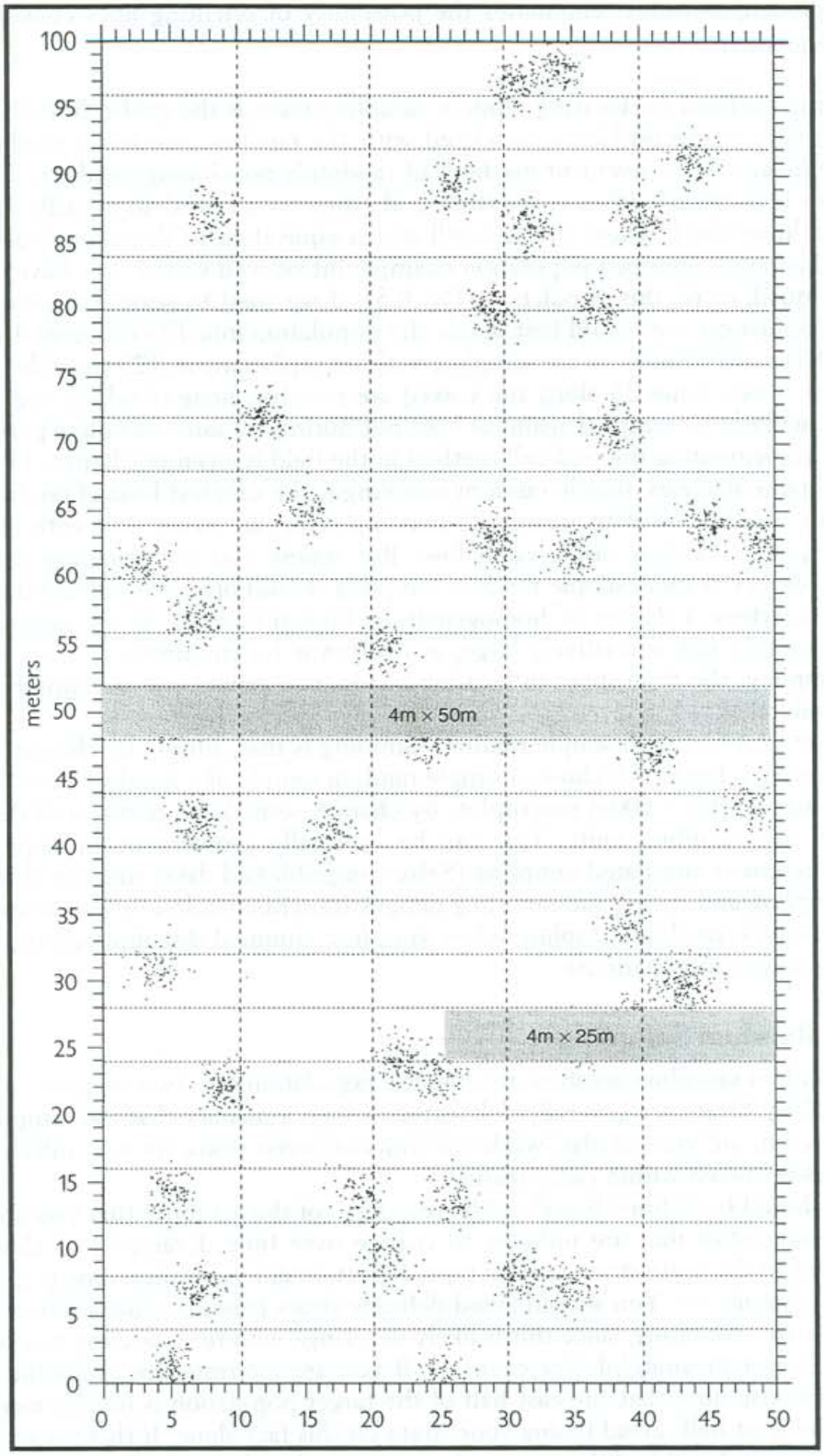

Figure 8.5. The dense clumped population overlaid with a grid of 4m × 10m quadrats. There are 125 possible quadrat locations for this size and shape of quadrat. The 4m × 25m quadrat (50 possible quadrat locations) and 4m × 50m quadrat (25 possible quadrat locations) are also shown. The 4m width was used for illustration only. A better quadrat design would be thinner (e.g., 0.25m or 0.5m) but would not show up well on the figure.

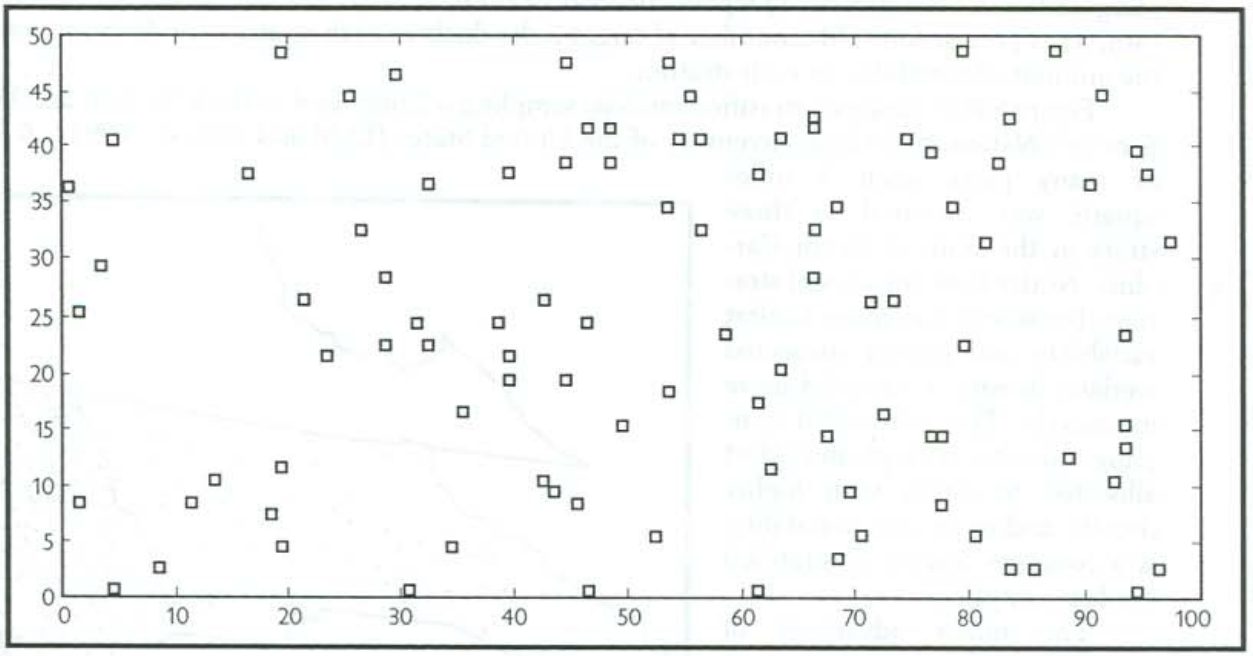

Figure 8.6. A simple random sample of 100 1m × 1m quadrats positioned within a 50m × 100m macroplot. Simply by chance, some large portions of the macroplot did not receive any sampling units.

random-sampling procedure, because you might find that your management will result in more recruitment of species X into the west half of the target population, leaving you with a stratified random sampling procedure that is less efficient than simple random sampling.

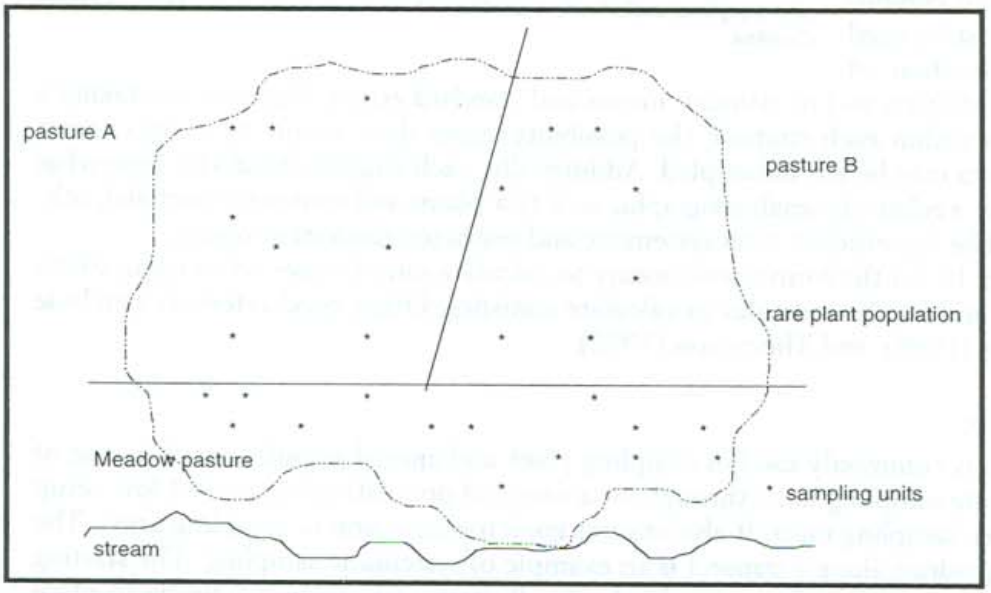

Figure 8.7. A rare plant population grows in a meadow along a stream and up an adjacent slope. The population area is grazed in the spring in Pasture A and in the fall in Pasture B. The meadow has recently been excluded from livestock grazing except for a short duration low intensity graze in the early spring before green-up. The three areas are treated as strata in a stratified random sample.

Figure 8.7 depicts a rare plant population that occurs within three grazing pastures, each with different grazing regimes. We decide to use each pasture as a sampling stratum. Through pilot sampling, we discover that the meadow portion of the population is more variable than the portion growing on the adjacent slope in the upland pasture, and we allocate more sampling units to that stratum.

Sampling units do not have to be allocated in equal numbers to each stratum. In fact, one of the benefits of stratified random sampling is that, when the attribute of interest responds differently to different habitat features, you can increase the efficiency of sampling over simple random sampling by allocating different numbers of sampling units to each stratum. Sam-

pling units can be allocated: 1) equally to each stratum, 2) in proportion to the size of each stratum, 3) in proportion to the number of target individuals in each stratum, or 4) in proportion to the amount of variability in each stratum.

Figure 8.8 illustrates a stratified random sampling scheme used in the U.S. Fish and Wildlife Service's National Wetlands Inventory of the United States (Dahl and Johnson 1991). A sample of many plots, each 4 miles square, was allocated to three strata in the state of North Carolina. Notice how the coastal stratum, because it has more habitat variability and greater suspected wetland density, is sampled more intensively. This differential sampling intensity, with greater effort allocated to strata with higher density and/or greater variability, is a common feature of stratified random sampling.

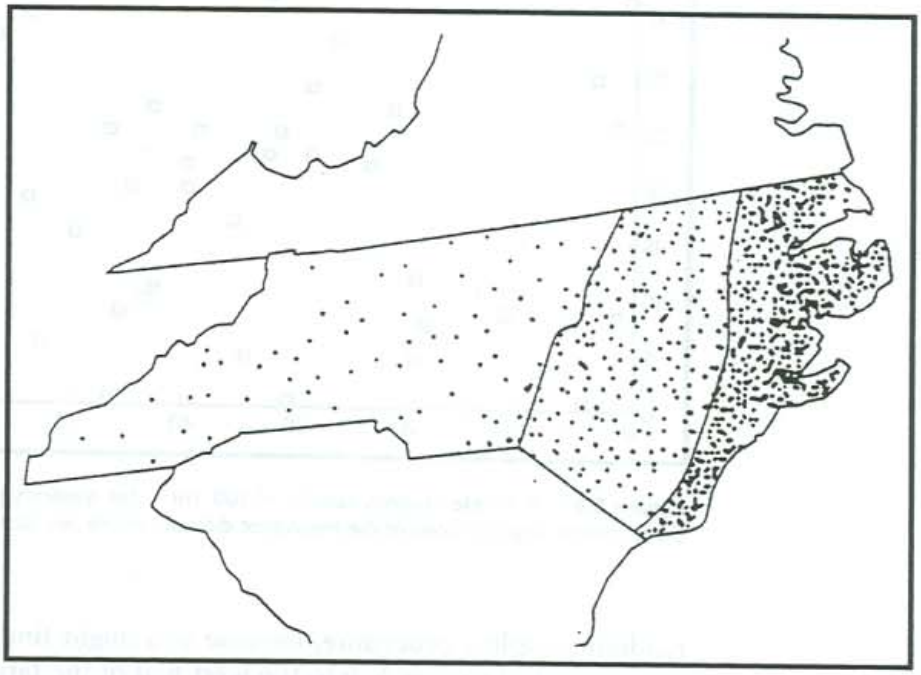

Figure 8.8. A stratified random sampling scheme. This example, from the National Wetlands Inventory (Dahl and Johnson 1991), shows how a sample of many plots, each 4 mi^2, was allocated to three strata in the State of North Carolina.

The major advantage of stratified random sampling is an increase in the efficiency of population estimation over simple random sampling when the attribute of interest responds very differently to some clearly defined habitat features that can be treated as strata. The principal disadvantage is the more complicated formulas that must be used both to determine allocation of sampling units to each stratum and to estimate means and standard errors. Since we are taking a simple random sample within each stratum, the possibility exists that, simply by chance, areas within one or more strata may be left unsampled. Additionally, each stratum should be somewhat homogeneous and cover a relatively small geographic area (for plants and stationary animals); otherwise the method will be less efficient than systematic and restricted random sampling.

Refer to Appendix IV for the formulas necessary to calculate sample sizes when using stratified random sampling and for the formulas to calculate statistics. Other good references include Cochran (1977), Krebs (1998), and Thompson (1992).

Systematic Sampling

A systematic approach is commonly used in sampling plant and animal populations. It is one of the easiest ways to locate sampling units throughout a sampled population because of low setup and travel time between sampling units. It also ensures good interspersion of sampling units. The regular placement of quadrats along a transect is an example of systematic sampling. The starting point for the regular placement is selected randomly. To illustrate, let us say we decide to place ten $1m^2$ quadrats at 5m intervals along a 50m transect. The selection of the starting point for systematic sampling must be random. Therefore, we randomly select a number between 0 and 4 to represent the starting point for the first quadrat along the transect and place the remaining nine quadrats at 5m intervals from this starting point. Thus, if we randomly select the 3m mark for the first quadrat, the remaining quadrats will be placed at the 8, 13, 18, 23, 28, 33, 38, 43, and 48m points along the transect. This is illustrated in the transect along the left side of Figure 8.9.

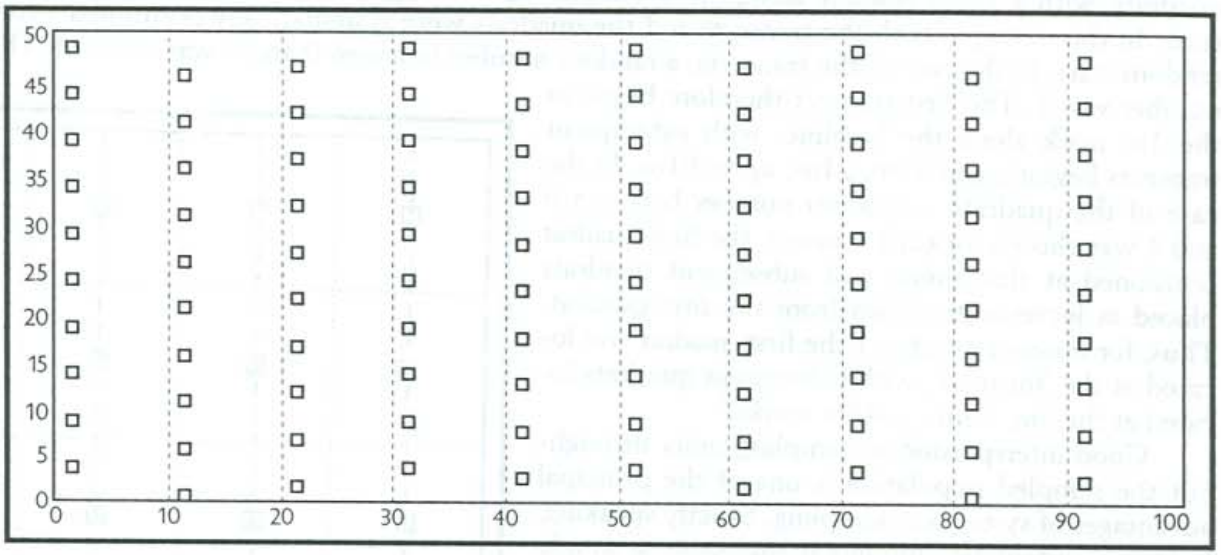

Figure 8.9. A 50m × 100m macroplot, sampled by 100 1m × 1m frequency quadrats. The quadrats are aligned along transects. Both the transects and the quadrats are systematically positioned with a random start. A random starting point is selected for the transects along the baseline, while separate random starting points are selected for the quadrats along each transect.

Systematic sampling with a random starting point is commonly used in animal studies because it permits easily identifying sampling points and because it generally, but not always, yields estimates of comparable accuracy and precision to those provided by purely random sampling. For example, for sampling fishes in small streams, a systematic sampling approach has been recommended (Hankin and Reeves 1988) because it delivers comparable precision, is generally representative, and avoids the work of identifying a complete list of sampling sites required by random sampling. Litter searches of quadrats for amphibians and small lizards are also frequently made in a systematic fashion. Similarly, point counts for birds are almost invariably arrayed in a systematic fashion along counting "routes" or transects.

A common use of systematic sampling in vegetation studies is to facilitate the positioning of quadrats for frequency sampling and of points for cover estimation. Using this approach, a baseline is laid across the sampled population, either through its center or along one side of it. Transects are run perpendicular to the baseline beginning at randomly selected points along the baseline (if the baseline runs through the middle of the population, transects are run in either of two directions; the direction for each one can be randomly determined by tossing a coin). Quadrats or points are then systematically positioned along each transect. The starting point for the first quadrat or point along each transect is selected randomly.

Systematic samples, if well designed, can safely be analyzed as a simple random sample. Milne (1959) analyzed data taken from random and systematic samples of 50 totally enumerated biologic populations and found that there was no error introduced by assuming that a centric systematic sample is a simple random sample and by using all the appropriate formulas from random sampling theory (Krebs 1998:228). Milne's (1959) conclusion was that "with proper caution, one will not go very far wrong, if wrong at all, in treating the centric systematic-area sample as if it were random." Note, however, that Milne compared random samples to centric systematic samples, illustrated in Figure 8.10. The units of a centric systematic sample lie on equidistant parallel lines (these can be thought of as transects) arranged in a manner such that, in effect, the area is divided into equal squares (see dotted lines) and a sampling unit taken from each square. Thus, the sampling units are spaced a considerable distance apart with maximum interspersion of sampling units throughout the sampled population.

The design shown in Figure 8.9 ensures good interspersion of sampling units throughout the sampled population. Here, a 50m × 100m macroplot was sampled by a hundred $1m^2$ frequency

quadrats, with a 100m baseline along the southern edge. The quadrats were aligned along transects. In this example both the transects and the quadrats were systematically positioned with a random start. In the case of the transects, a random number between 0 and 9 was selected. That number was 1. The first transect therefore began at the 1m mark along the baseline, with subsequent transects beginning at 11m, 21m, up to 91m. In the case of the quadrats, a random number between 0 and 4 was chosen for each transect, the first quadrat positioned at that point, and subsequent quadrats placed at increments of 5m from the first quadrat. Thus, for transect number 1 the first quadrat was located at the 3m mark, with subsequent quadrats located at the 8m, 13m . . . 48m marks.[4]

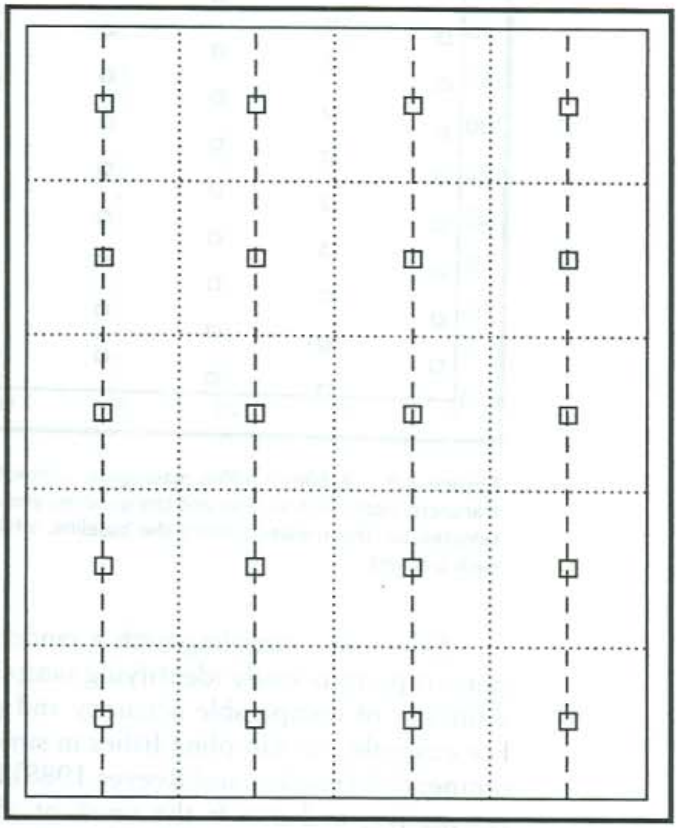

Figure 8.10. A centric systematic sample (adapted from Milne 1959). Small squares are sampling units, dashed lines are transects, and dotted lines show how the sampling units fall in the center of each subunit of area.

Good interspersion of sampling units throughout the sampled population is one of the principal advantages of systematic sampling. Strictly speaking, however, systematic sampling is analogous to simple random sampling only when the population being sampled is in random order (see, for example, Williams 1978). Populations in random order are rare in biology; most natural populations of both plants and animals exhibit a clumped spatial distribution pattern. This means that nearby units tend to be similar to (correlated with) each other. If, in a systematic sample, the sampling units are spaced far enough apart to reduce this correlation, the systematic sample will tend to furnish a better mean and smaller standard error than is the case with a random sample, because with a random sample one is more likely to end up with at least some sampling units close together (see Milne 1959; discussion of sampling an ordered population in Schaeffer et al. 1979). Computer simulation has validated this conclusion. For example, for density estimation, Salzer (unpublished data) found through Monte Carlo simulations that systematic designs outperform simple random sampling in terms of precision when sampling clumped populations.

On a cautionary note, systematic sampling for density estimation can lead to questionable results if the sampling design creates a situation where there are only a small number of potential samples. For example, consider the macroplot shown in Figure 8.11. Ten 1m × 50m quadrats are systematically positioned in the macroplot with a random starting point at the 2m position on the x-axis, and the quadrats spaced at 10m intervals after that. In this case, since the position of all quadrats is fixed once the first quadrat is positioned, there are only 10 possible samples to draw from, depending on which of the 10 possible starting points is randomly selected in the first 10m segment of the population (0, 1, 2, 3, 4, 5, 6, 7, 8, or 9). The sampling distribution (distribution of all possible sample mean values) for this sampling design might resemble a uniform (flat) distribution instead of the smooth, bell-shaped curve of the normal distribution, because

[4]*What is the sampling unit in Figure 8.9? You have two options: You can treat the sample as if the quadrats had been selected as a simple random sample or you can calculate separate frequency values for each transect and treat the transect as the sampling unit. The implications of each option will be clearer once you have been introduced to cluster sampling and two-stage sampling, discussed below.*

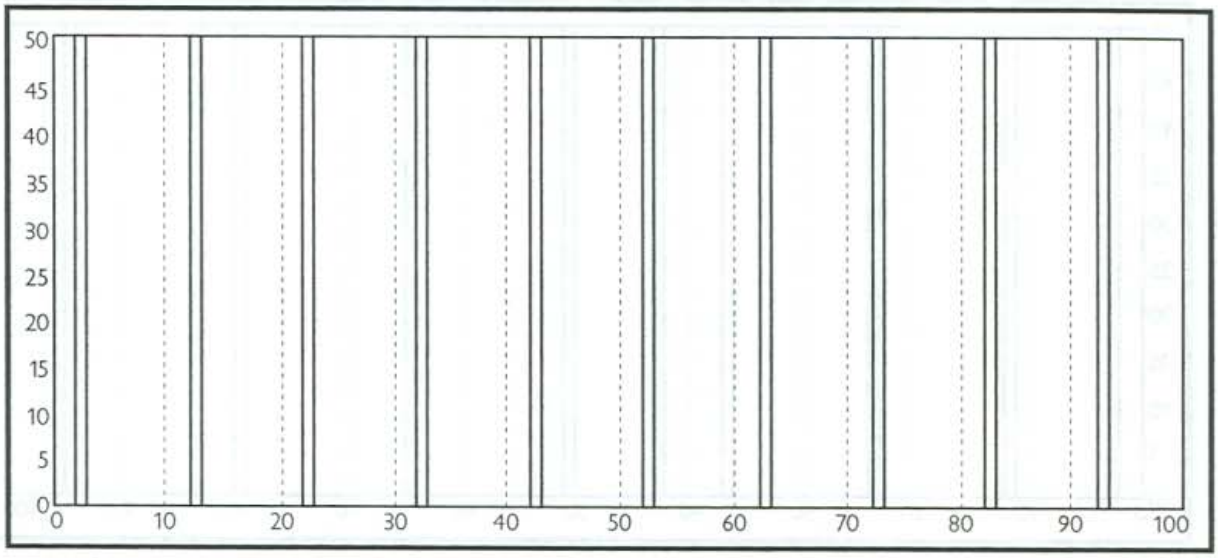

Figure 8.11. A systematic sample of 10 1m × 50m quadrats in a 50m × 100m macroplot. Note that there are only 10 possible samples, corresponding to which of the 10 possible starting points in the first 10m segment of the baseline (x-axis). In this case, the sample started at the 2m mark.

there are only 10 different sample means possible. Treating such a sample as if it were a simple random sample could result in poor estimates of the sample standard error. The next type of sampling design, restricted random sampling, solves this problem. Except for this somewhat uncommon situation, however, systematic sampling is preferred over restricted random sampling. If more than 30 possible systematic samples may be drawn, systematic sampling is acceptable.

Another caution is that situations do arise in which systematic sampling can seriously bias estimates if the pattern of the sampling units intersects some pattern in the environment (e.g., dune ridges and slacks; Goldsmith et al. 1986). One example is estimating food abundance for wildlife in croplands planted in a regularly repeated fashion. Systematic sampling, depending on how it was applied, might consistently locate sampling units between or on top of crop rows and thereby yield substantially different estimates. This has occurred, for example, in studies of availability of waste corn for waterfowl.

If some periodic pattern does exist, the data analysis will not reveal this, and your estimates, particularly of standard errors, will be wrong. Although this type of periodic pattern is rare in nature, you should be alert to the possibility.

Restricted Random Sampling

In restricted random sampling, you determine the number of sampling units, n, you will need to meet your monitoring objective (sample size determination is discussed below), then divide your population into n equal-sized segments. Within each of these segments, a single sampling unit is randomly positioned. The sample of n sampling units is then analyzed as if it were a simple random sample.

Figure 8.12 is an example of a restricted random sampling procedure. This is the same 50m × 100m macroplot as we used in our discussion of systematic sampling. In this case, however, we divide the x-axis into ten 10m segments. Within each of these segments we randomly select a single quadrat location. This gives us 10 possible random locations within every 10m segment of the x-axis. Every quadrat location in the macroplot still has an equal probability of selection. The same technique can also be applied to the y-axis if there is more than one possible quadrat position along that axis.

The restricted-random-sampling procedure can also be used when the sampling unit is a transect instead of a quadrat. Divide the population into equal-sized segments and allocate a single transect to each segment. If you are locating sampling units such as quadrats or point intercepts along transects (similar to Figure 8.9), you may want to use a combination of the restricted

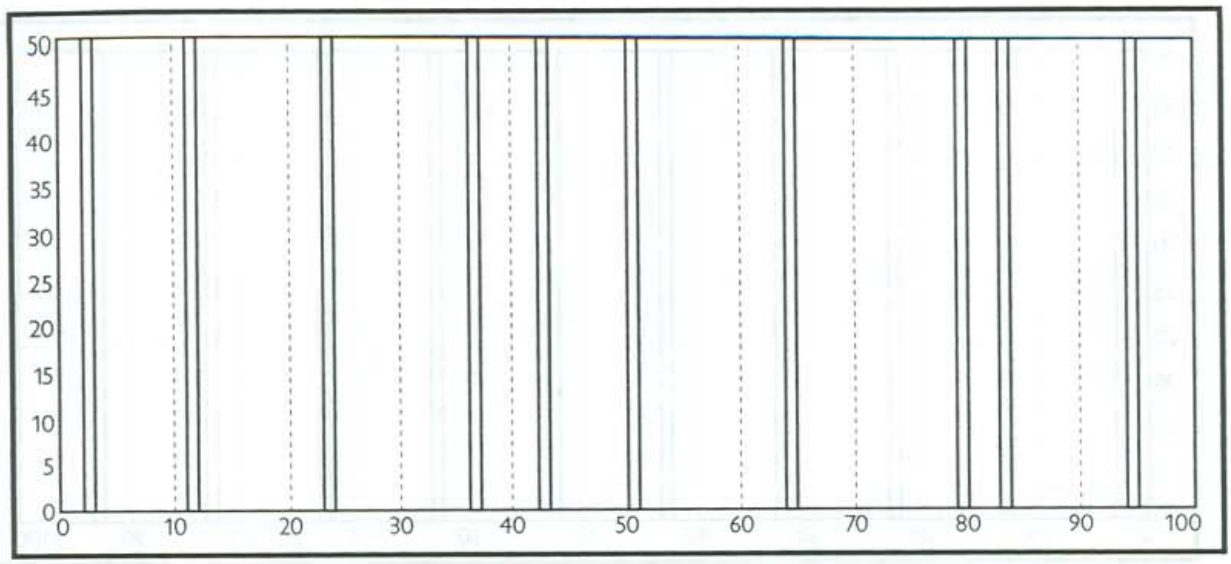

Figure 8.12. A restricted random sample of 10 1m × 50m quadrats in a 50m × 100m macroplot. One quadrat is randomly positioned within each 10m segment of the baseline (x-axis).

and systematic designs. If, for example, you decide to run 10 transects, each with 50 point intercepts, perpendicular in one direction from a baseline, you could divide the baseline into 10 equal segments, randomly locate beginning points for each transect within each of these 10 segments, and then systematically space the point intercepts along each transect (as in Figure 8.11, except with points systematically positioned along one edge of each quadrat).

Restricted random sampling is similar to both stratified random and systematic sampling. It is similar to stratified random sampling in that we have effectively stratified our macroplot into 10 strata. However, unlike stratified random sampling, the strata are arbitrary, and we take only one sampling unit in each stratum. As with systematic sampling, we divide our population into equal-sized segments. With systematic sampling, however, only the first sampling unit is randomly determined; all the others are spaced at equal intervals from the first.

Similar to systematic sampling, restricted random sampling results in very good interspersion of sampling units throughout the target population. Furthermore, Salzer (unpublished data) has shown through simulation studies that restricted random sampling results in more precise estimates of density than simple random sampling. He has also demonstrated the procedure to be more robust than systematic sampling when the number of possible systematic samples are few, because with restricted random sampling designs you do not constrain the number of potential samples from which you can draw. The principal disadvantage of restricted random sampling is that you can, purely by chance, end up with sampling units positioned side-by-side. This can leave larger portions of the sample area unsampled than is the case with a systematic design. When the number of potential systematic samples is large enough (more than 25 to 30), you are probably better off choosing a systematic sample. Otherwise, use the restricted random design.

Cluster Sampling

Cluster sampling[5] is a method of selecting a sample when it is difficult or impossible to take a random sample of the individual elements of interest. With cluster sampling, we identify groups or clusters of elements and take a random sample of these clusters. We then measure every element within each of the randomly selected clusters.

[5]*Cluster sampling should not be confused with cluster analysis, a technique used in classification and taxonomy.*

In monitoring, cluster sampling is most often used when the objective is to estimate something about individuals such as parasite loads in animals or the mean number of flowers per plant. For example, you may want to track the average height of plant X in population Y. There are too many plants in the population to feasibly measure all of them. Five quadrats are randomly placed in the population, and the heights of all plants within these quadrats are measured (Fig. 8.13).

Cluster sampling and two-stage sampling are the only two efficient designs that can be used to sample individual plant and animal characteristics. Examples for application to plant monitoring include estimating number of seeds produced per plant, biomass per plant, and average height or size per plant. In these examples, a quadrat is employed as the cluster and each plant is an element. Examples of the application of two-stage and cluster sampling to animal studies include estimating the size of birds' eggs (nests are the cluster and eggs are the elements), the number of eggs per nest (nests may be located using trees or a quadrat as the cluster and nests as the element), food habits of fish (e.g., a seine catch as the cluster and each fish stomach as an element), and the size of beetles (the trap is the cluster and each beetle the element). Sometimes,

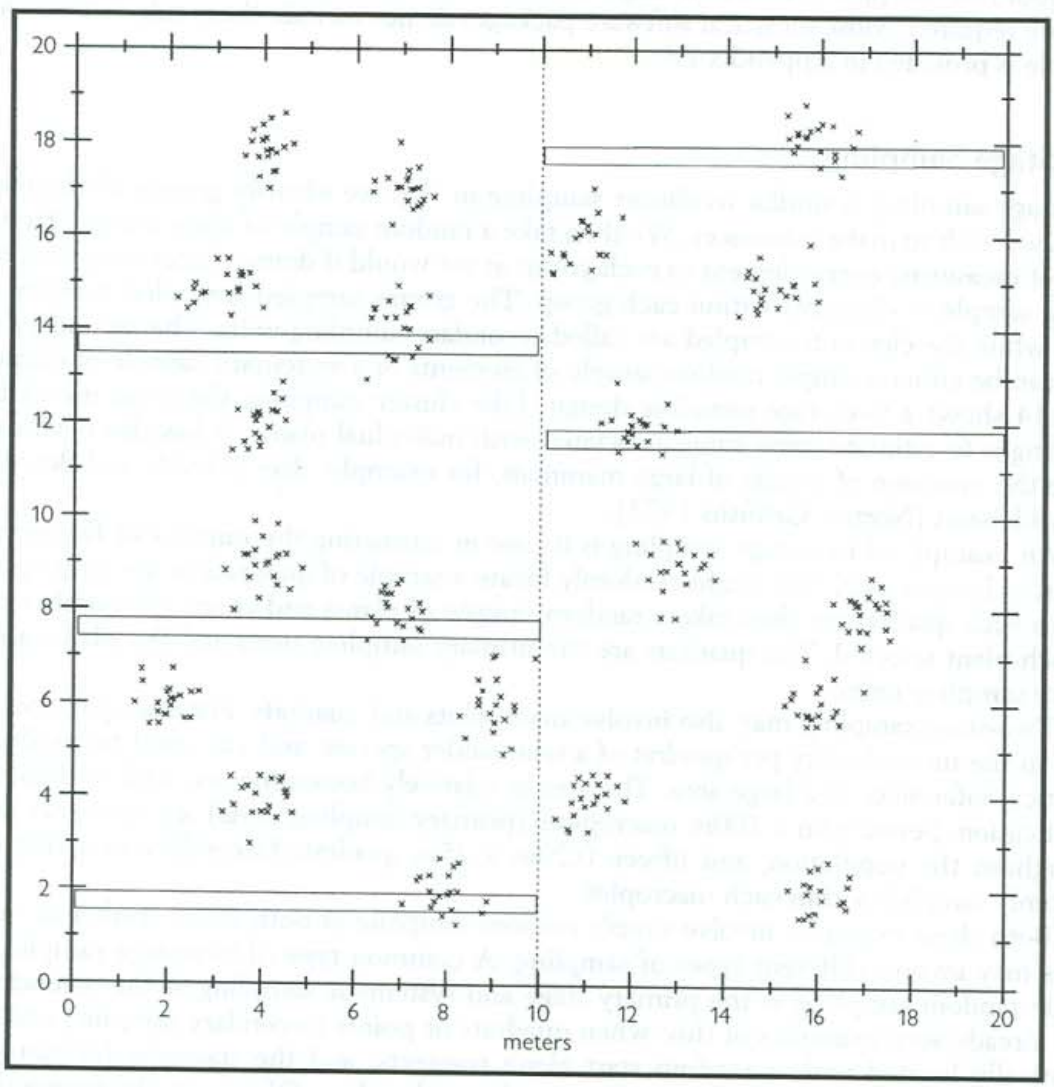

Figure 8.13. An example of cluster sampling to estimate the mean height of plants in a population. Five quadrats are randomly placed in the population and the heights of all plants within these quadrats are measured.

the elements are erroneously treated as independent sampling units. Careful articulation of the method of positioning sampling units should help avoid this problem.

With animals, cluster sampling is also sometimes of a temporal rather than a spatial nature, such that repeated counts are made during randomly determined visits to a site instead of making the single counts at randomly determined times, thereby greatly saving on time needed to reach sites to make counts.

The advantage of cluster sampling is that it is often less costly to sample a collection of elements in a cluster than to sample an equal number of elements selected at random from the population (Thompson 1992). It is most efficient when different clusters are similar to each other and incorporate much variability within. Because individuals near each other tend to be similar, this condition will not be realized with square clusters (Thompson 1992). Therefore, just as with simple random sampling for density estimation, cluster sampling using long, narrow quadrats to delineate clusters will be more efficient than using square quadrats.

Cluster sampling has several disadvantages. First, all elements within each cluster must be measured. If the clusters contain large numbers of the element of interest, two-stage sampling, described below, will be more efficient. Second, it is often difficult to figure out how many clusters should be sampled versus how large each cluster should be. Third, more complex calculations are required. Most statistical software packages do not include these calculations. A worked example is provided in Appendix IV.

Two-Stage Sampling

Two-stage sampling is similar to cluster sampling in that we identify groups of elements about which we wish to make inferences. We then take a random sample of these groups. However, instead of measuring every element in each group as we would if doing cluster sampling, we take a second sample of elements within each group. The groups sampled are called primary sampling units, while the elements sampled are called secondary sampling units. The secondary sampling units can be either a simple random sample of elements or a systematic sample of elements. Figure 8.14 shows a two-stage sampling design. Like cluster sampling, the main use of two-stage sampling is to estimate some value associated with individual plants. It has also been used to increase the precision of counts of large mammals, for example, deer (Freddy and Bowden 1983) and wildebeest (Norton-Griffiths 1973).

An example of two-stage sampling is its use in estimating the number of flowers per plant produced by species X. We might randomly locate a sample of quadrats in the target population. Within each quadrat we then take a random sample of plants and count the number of flowers on each plant selected. The quadrats are the primary sampling units and the plants are the secondary sampling units.

Two-stage sampling may also involve macroplots and quadrats. For example, you are interested in the mean density per quadrat of a salamander species, and you want to be able to make statistical inferences to a large area. The area is relatively homogeneous, with no logical basis of stratification. Seven 50m × 100m macroplots (primary sampling units) are randomly distributed throughout the population, and fifteen 0.20m × 25m quadrats (secondary sampling units) are randomly sampled within each macroplot.

Both these examples involve simple random sampling at both stages. Either or both of the stages may involve different types of sampling. A common type of two-stage sampling involves simple random sampling at the primary stage and systematic sampling at the second stage. We have already seen examples of this: when quadrats or points (secondary sampling units) are systematically located with a random start along transects, and the transects (primary sampling units) are run from randomly selected points along a baseline. Of course, the transects could be positioned using another type of design such as restricted random sampling or systematic sampling. The point is that the two stages can involve different sampling designs.

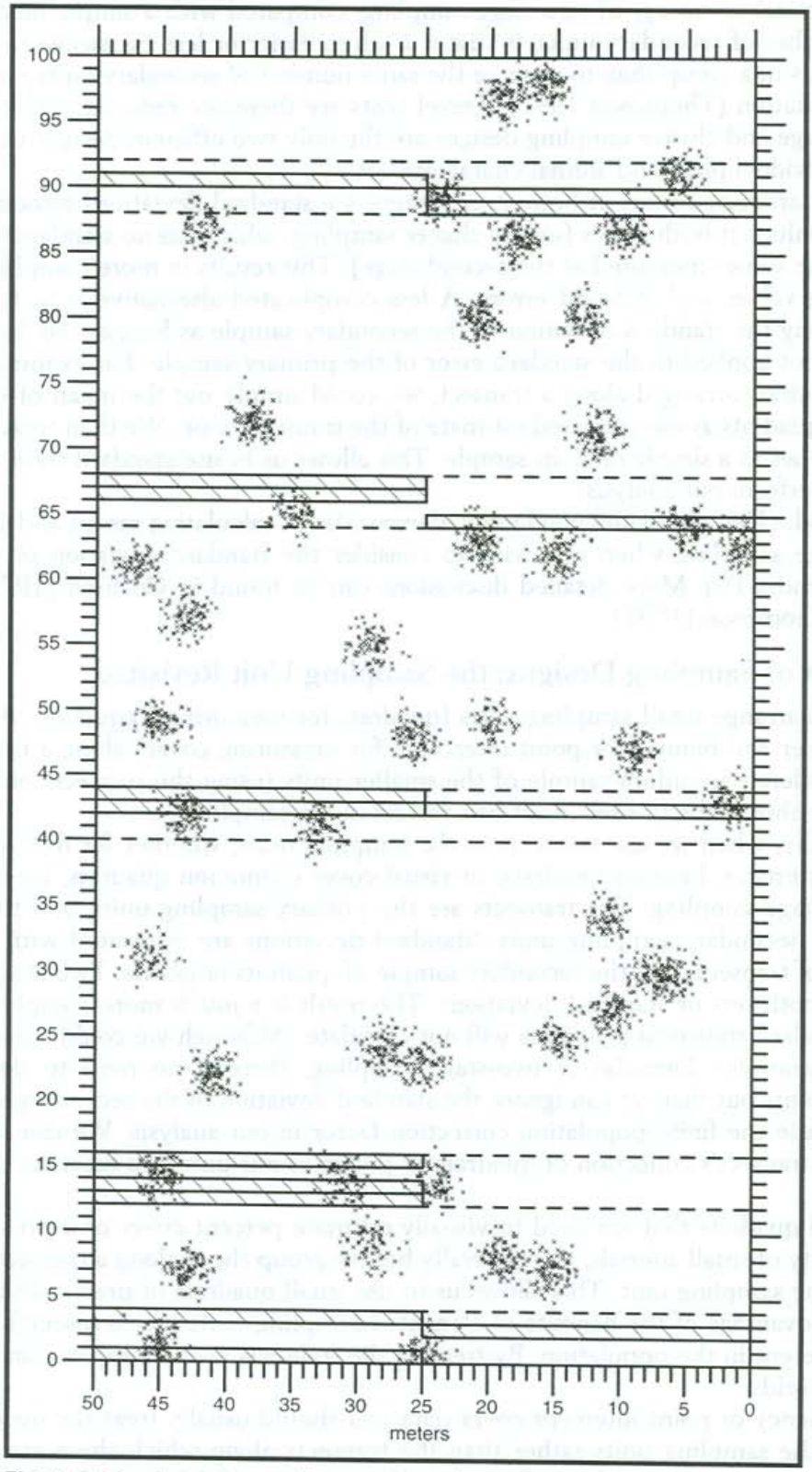

Figure 8.14. Two-stage sampling to estimate the number of flowers per plant on a particular species of plant. Five 4m × 50m quadrats (primary sampling units) are randomly located in the sampled population and three 1m × 25m quadrats (secondary sampling units) are randomly located within each of the five larger quadrats. The number of flowers per plant is counted within all of the selected 1m × 25m quadrats.

The practical advantage of two-stage sampling, compared with a simple random sample of the same number of secondary units, is that it is often easier or less expensive to observe many secondary units in a group than to observe the same number of secondary units randomly spread over the population (Thompson 1992). Travel costs are therefore reduced with two-stage sampling. Two-stage and cluster sampling designs are the only two efficient designs that can be used to sample individual plant and animal characteristics.

Because sampling occurs at both stages, there are standard deviations associated with estimates of the values at both stages (unlike cluster sampling, which has no standard deviation associated with the values measured at the second stage). This results in more complicated formulas for estimating values and standard errors. A less complicated alternative is to follow Cochran (1977), ignoring the standard deviation of the secondary sample as long as the finite population correction is not applied to the standard error of the primary sample. For example, if we had a sample of quadrats arranged along a transect, we could simply use the mean of each transect's collection of quadrats as our unbiased estimate of the transect value. We then treat the collection of transect values as a simple random sample. This allows us to use standard statistical computer programs to perform our analysis.

Platts et al. (1987) provides good worked examples of calculating means and standard errors from two-stage sampling when you wish to consider the standard deviation of the secondary sample (Appendix IV). More detailed discussions can be found in Cochran (1977:279), Krebs (1998), and Thompson (1992).

Comparison of Sampling Designs: the Sampling Unit Revisited

Often we will arrange small sampling units (quadrats for measuring frequency, visual estimates of percent cover and biomass or point intercepts for measuring cover) along a transect. Should these be considered a random sample of the smaller units (using the transects only for locating these units) or should the transect itself be considered the sampling unit?

Technically, when we use transects as the sampling units, whether for frequency quadrats, cover point estimates, biomass quadrats, or visual cover estimation quadrats, we are really conducting two-stage sampling. The transects are the primary sampling units, and the quadrats or points are the secondary sampling units. Standard deviations are associated with both the primary sample of transects and the secondary sample of quadrats or points. Two-stage designs take into account both sets of standard deviations. The result is a much more complex set of equations that standard statistical programs will not calculate. Although we could subject these data to the more complex formulas of two-stage sampling, there is no need to do so. Cochran (1977:279) points out that we can ignore the standard deviation of the secondary sample as long as we do not use the finite-population correction factor in our analysis. We can simply use the mean of each transect's collection of quadrats or points as our unbiased estimate of the transect value.

For small quadrats that are used to visually estimate percent cover or estimate biomass or estimate density of small animals, it is generally best to group those along a transect and consider the transect the sampling unit. This allows us to use small quadrats of practical size in the field while taking advantage of the benefits of elongated sampling units (the transects) that cross the variability inherent in the population. By treating the transects as the sampling units, we get the best of both worlds.

For frequency or point intercept cover data you should usually treat the quadrats or point intercepts as the sampling units rather than the transects along which these are located. Estimates will be more precise and significance tests more powerful because of the larger sample sizes realized by using quadrats or point intercepts rather than transects as the sampling units. There are at least two situations, however, in which you might want to treat the transects as the sampling units. The first of these is when the transects are permanent (see discussion on permanent vs. temporary sampling units below). If you have reason to believe that the average values

per transect are more correlated between years than are the quadrat or point values, you may choose to analyze the transects rather than the quadrats or points as the sampling units.[6]

The second situation in which you might want to treat the transects as the sampling units when systematically sampling frequency quadrats or cover point intercepts is when the quadrats or points are not far enough apart to be considered independent. This is more likely to be a problem in already established studies, where quadrats or points were placed contiguously or a very short distance apart. Hopefully, you will design new studies in such a manner that the quadrats or points are spaced far enough apart to achieve independence.

Independence means that the sampling units are not spatially correlated, that the response of the species in Quadrat A is not related to the response of the species in Quadrat B because of their proximity to one another. If the quadrats or points are far enough apart that they can be considered independent, we have the benefit of increasing our sample size dramatically (because the point or plot is the sampling unit instead of the transect) while keeping the field efficiency of locating sampling units rapidly along a transect. Conversely, the contiguous placement of quadrats along a transect or the separation of such quadrats by small distances (e.g., one "pace"), practically ensures that adjacent sampling units will be correlated. This will result in an underestimation of the standard error and questionable results

Determining how far apart to place sampling units along a transect for them to be considered independent can be difficult. Chapter 12 discusses this issue in more detail for plants. This is a particular problem for animals as well, especially those that are detected at long distances by their calls and hence easily double-counted. These issues are discussed in Chapter 13. Probably the best way to determine spacing of sampling units along transects is to consider the degree of interspersion of your design. The goal is to have sampling units interspersed as well as possible throughout the area of the target population (see previous discussion on interspersion). Once you have delineated the area you intend to sample, strive for a design in which the spacing between transects is about the same as the spacing between sampling units. If you do this, it is likely that the issue of independence will take care of itself.

Double Sampling

Double sampling, sometimes called two-phase sampling, involves the estimation of two variables. Because one of these variables, the variable of interest, is difficult and expensive to measure, it is measured in only a relatively small number of sampling units. To improve the rather poor precision of the estimate that normally results from a small sample, an auxiliary variable that is much easier to measure is estimated in a much larger number of sampling units. The variable of interest is measured in a subsample of the sample of units in which the auxiliary variable is measured.

The idea of double sampling will become clearer with examples. The technique is often used in estimating aboveground biomass in rangelands. Because it is slow and expensive to clip, dry, and weigh biomass in many sampling units, observers train themselves to visually estimate biomass. Once trained, the observers randomly locate quadrats within a target population and visually estimate the biomass in all the quadrats. For example, 100 quadrats are so estimated. Then, in a subsample of these quadrats, say 10, the visual estimates are made as in the other quadrats, but after these estimates are recorded, the aboveground biomass is clipped, dried, and weighed. Thus, for these 10 quadrats we have two estimates of biomass, one from the visual estimate, the other from the actual weighing of the clipped biomass.

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[6] *It is highly unlikely that you will be able to accurately reposition point intercepts as permanent sampling units, but a transect of point intercepts may be highly correlated from year to year and thus be suitable for consideration as a permanent sampling unit.*

Double sampling is also used in forest surveys to estimate the volume of trees in a stand. Trained observers make a visual estimate of volume for a large sample of standing trees, while accurate volume measurements that require felling are limited to a small subsample of trees (Thompson 1992).

In wildlife management, double sampling may be used for surveys of breeding waterfowl. Aerial surveys estimate abundance over an extensive area, but a subsample of the survey areas are subjected to more thorough ground surveys. The ground surveys are used to adjust the bias inherent in aerial surveys (Routledge 1999).

In all these cases, the subsample on which the variable of interest is actually measured is more accurate, but the precision of the estimate can be greatly improved by considering the measurements on the auxiliary variable. The improvement in precision depends on how well the auxiliary variable correlates with the variable of interest. In the examples given above, this relates to how well the trained observers actually estimate biomass, tree volume, or abundance of waterfowl.

If the auxiliary variable is relatively quick to be measured and is highly correlated with the variable of interest, double sampling is much more efficient in estimating variables that are difficult to measure compared to directly measuring the variable in all sampling units. A disadvantage is that the formulas for data analysis and sample-size determination are much more complicated than formulas for simple random sampling. Refer to Cochran (1977:327-358) or Thompson (1992:139-147) for the formulas needed to analyze double-sampling data.

Taking a Simple Random Sample of Individual Plants or Animals

Let us say that we want to estimate something about a population of individual plants such as their mean height or the mean number of flowers per plant, and that the population is too large to measure this variable on every single plant in the population. Easy, you say; we will just take a simple random sample of plants, measure the variable on the sample, and calculate the mean and standard error for the sample. We can then construct a confidence interval around the estimate at whatever confidence level we choose (e.g., a 95% confidence interval). Although it might seem logical to take a simple random sample of plants, for most plant populations this is not feasible.

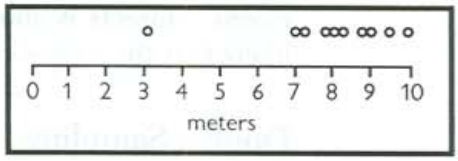


Figure 8.15. Distribution of individuals of plant species X along a 10-meter transect. A randomly positioned point on the transect will be far more likely to be closest to the individual at the 3m mark than to any of the other plants.

One way that is often—and incorrectly—used is to select a random sample of points in the population and to take the nearest individual to each of these points. Unfortunately, this works only if the population of plants or animals is randomly distributed, a condition rarely met by natural populations. If, as is typically the case, the individuals are spatially distributed in patches, this technique most decidedly will not result in a simple random sample of individuals. Consider Figure 8.15, which shows the distribution of individuals of a hypothetical plant species along a 10m transect. Note that nine of the ten individuals are clumped in the last 3 meters of the transect, while a single individual occurs at the 3m mark. A randomly positioned point along this transect would have about a 50% probability of being closest to this isolated individual and about a 20% chance of being closest to the individual at the 7m mark. The probability of the point lying closest to any of the other eight individuals is much less.

Thus, in a clumped population of plants, a "random" sample of individuals chosen by taking the individuals closest to randomly located points will be biased toward those individuals that are isolated from the majority of the population. These individuals may either be much larger than the majority of plants in the population because of reduced intraspecific competition or much smaller because they occupy suboptimal habitat. Let us say we are interested in estimating the mean height of such a population. By biasing our estimate toward the isolated plants in the population we may greatly underestimate or overestimate the mean height of such

a population. The same is true for any other attribute associated with individual plants that we may wish to estimate such as number of fruits per plant. Obviously, for populations of plants that follow a clumped or patchy distribution—which is by far the majority of populations—such a sample of individuals cannot be used to adequately characterize the population.

How, then, can you take a random sample of individuals? One way is to completely enumerate every individual in the population by, for example, mapping every individual and numbering each one from 1 to *n*. A simple random sample could then be taken by drawing random numbers between 1 and *n*. This, of course, would be extremely time-consuming except for small populations, in which case you might be able to measure the attribute on every individual in less time. For example, if you are interested in mean height of plants, you could simply measure the height of every plant in the population and not sample at all. If, however, you need to estimate the mean number of flowers per plant and each plant has several hundred flowers, selecting individual plants randomly from a complete list might be a reasonable approach, although for most practical purposes it is far too time-consuming.

Another possibility is to take a systematic random sample of individuals. With this method you gather information from every *nth* individual in the population. This method will work if you are planning to conduct a complete census of the population, but you are also interested in estimating some attribute from a subset of the individuals (e.g., number of flowers/plant). Before you start you need an estimate of the following two types of information: 1) the approximate size of the population, and 2) the approximate number of individual plants you will need to sample (calculated as a proportion of the total population size). For example, if you estimated a total population size of 1000 plants and your sample-size calculations from pilot sampling identified a sample size of 100 plants, you would count the number of flowers on every 10th plant encountered. You choose a random number between 1 and 10. Say the number is 4. Then, starting at one edge of your population you systematically count the plants. You place a pin flag next to plant number 4, another next to plant number 14, and so on until you have counted all the plants. You can then come back and count the flowers on the flagged plants. This sample can properly be analyzed as a simple random sample.

The most practical approach to estimating attributes of individual plants and animals usually employs cluster sampling or two-stage sampling designs using quadrats as primary sampling units. In a cluster sample you would measure the attribute on all the plants in the primary sampling unit (the quadrat). If individuals are still too numerous within the quadrat to measure all of them, you could employ a two-stage sampling design by positioning smaller quadrats (secondary sampling units) within each large quadrat (primary sampling units).

SHOULD SAMPLING UNITS BE PERMANENT OR TEMPORARY?

A critical decision in sampling designs for monitoring is whether to make your sampling units temporary or permanent. When sampling units are temporary, the random sampling procedure is carried out independently at each sampling period. For example, your sampling objective involves detecting change in density over time of a plant species in a 50m × 100m macroplot. In the first year of sampling you place twenty-five 0.5m × 25m quadrats within the macroplot by randomly selecting 25 unique sets of coordinates and counting the number of the species in each quadrat. In the second year of sampling, you place another twenty-five 0.5m × 25m quadrats by randomly selecting a new set of coordinates and counting the number of the species in each quadrat. The sampling units (quadrats) in this example are temporary, and the two samples are independent of each other.

Using the same sampling objective, you could decide to use permanent quadrats. In the first year of sampling you randomly place the 25 quadrats as described above and count the number of individuals in each quadrat. This time, however, you permanently mark the locations of the 25 quadrats. In the second year of sampling, you count the number of individuals in the same quadrats. In this example the sampling units are permanent, and the two samples are dependent.

The principal advantage of using permanent instead of temporary sampling units is that for many species the statistical tests for detecting change from one period to the next in permanent sampling units are much more powerful than the tests used on temporary sampling units. This advantage translates into a reduction in the number of sampling units that must be sampled to detect a certain magnitude of change.

To see why this is so, let us consider the process used in comparing the samples between two periods when using permanent quadrats. If we were using temporary quadrats, we would calculate separate means and standard errors for the two samples and compare these using a statistical test (such as a *t*-test) for independent samples (see Chapter 9). With permanent quadrats, however, we calculate only one mean and one standard error. This requires some explanation. Each quadrat at time one is paired with the same quadrat at time two. The data from which we calculate the mean and standard error consists of the set of differences between each of the quadrats at time one and its corresponding quadrat at time two. For example, we randomly positioned five permanent quadrats in a population and counted the number of plants in each quadrat in 1993 and again in 1994. Data from these permanent quadrats yielded the values in Table 8.2.

Quadrat Number	Number of Plants in 1993	Number of Plants in 1994	Difference Between 1993 and 1994
1	5	5	0
2	5	5	0
3	5	5	0
4	6	6	0
5	3	3	0
			mean difference 0
			standard error 0

Table 8.2. Density Data Taken From Five Permanent Quadrats in 1993 and 1994.

Note that the permanent quadrats are extremely effective at detecting the lack of change from year to year. Because in our example the difference between 1993 and 1994 was zero in every quadrat, there is no variation between sampling units, and the standard error is actually 0. Had temporary quadrats been used in both years, it is quite likely that the estimates for each year would have been different just because of chance. For this reason more temporary sampling units (perhaps many more) would have been required to reach the same conclusion that no change had occurred.

Because we are interested only in the change that takes place within each permanent sampling unit between two periods, the difference between sampling units at either period is not nearly as important as it is when using temporary quadrats. Consider the following example. To detect change in cover of species X between two periods, 10 transects were randomly positioned in the target population in 1990. The beginning, middle, and end points of each transect were permanently marked. Fifty points were systematically positioned (with a random start) along each transect and "hits" recorded on canopy cover of species X. The estimate of cover along each transect is then this number of hits divided by the total number of possible hits, 50. Thus, a transect with 34 hits would have a cover estimate of 68% or 0.68. The data from these two years are shown in Table 8.3. (This example is also displayed graphically in Figs. 9.10 and 9.11 of Chapter 9.)

Even though the cover estimates are highly variable between transects for both 1990 and 1994 (for example the mean cover for 1990 is 0.44 with a 95% confidence interval of 0.27 to 0.62), the standard error of the mean difference is relatively small. A 95% confidence interval around this mean difference is –0.02 to –0.12. In fact, in lieu of doing a paired statistical test (such as a paired *t*-test), you could simply look at the 95% confidence interval around the mean difference to see if it includes 0. If not, then you can declare the change significant at a *P* value of 0.05 (*P* values are explained in Chapter 9).

If you had collected these data using temporary transects (i.e., independent samples at both sampling periods), you would have concluded that no change took place. In fact, with the large

Transect Number	Cover in 1990	Cover in 1994	Difference Between 1990 and 1994
1	0.22	0.20	-0.02
2	0.32	0.26	-0.06
3	0.06	0.06	0.00
4	0.86	0.80	-0.06
5	0.62	0.58	-0.04
6	0.54	0.50	-0.04
7	0.50	0.32	-0.18
8	0.28	0.24	-0.04
9	0.36	0.18	-0.18
10	0.68	0.64	-0.04
			Mean difference -0.07
			Standard error 0.02

Table 8.3. Cover Values Taken Along 10 Permanent Transects of 50 Points Each in 1990 and 1994.

degree of variability between transects, you would have needed unreasonably large numbers of transects to detect the change that only 10 permanent transects were able to detect.

This simple comparison suggests that permanent sampling units would always be advantageous, but their value must be balanced against their disadvantages. Time and equipment costs associated with permanent sampling units are higher than temporary ones. Sampling units must be marked well with permanent markers. These can be costly and time-consuming to install during the first year and difficult to find on subsequent years. Permanent markers may not be feasible in some situations because of the nature of the habitat or for safety reasons (see Chapter 5).

Another disadvantage of a design using permanent sampling units is that you usually need 2 years of data to determine adequate sample size. The only exception to this is when you have some basis to estimate the degree of correlation (the correlation coefficient) of sampling units between years when estimating means (e.g., density sampling) or a model of how the population is likely to change when estimating proportions (e.g., frequency sampling). We will discuss this at more length in the next section.

Impacts either from investigators or from animals may bias your results. By going back to the same sampling unit locations each year, you might negatively impact the habitat in or near the permanent sampling units. In addition, permanent markers may also attract wildlife, domestic livestock, wild horses, or burros. This might lead to differential impacts to the vegetation in or near the sampling units. If markers are too high (e.g., t-posts or other fence posts), livestock may use the markers for scratching posts and differently impact the sampling units. Wildlife impacts may also occur. Raptors, for example, might use the markers as perches; this could result in fewer herbivores in the sampling units than elsewhere in the target population, with resulting differences in the attribute being measured. Songbirds also might use the perches, defecating seeds and changing the plant community.

The advantage of permanent sampling units varies depending on degree of correlation between two measurements. Permanent sampling units will be the most advantageous when there is a high degree of correlation between sampling-unit values between two periods. This condition often occurs with long-lived plants (e.g., trees, shrubs, large cacti, or other long-lived perennial plants and long-lived and relatively sedentary animals). If, however, there is low correlation between sampling units between two periods, then the advantage of permanent quadrats is diminished. This could occur, for example, with annual plants, if their occurrence in quadrats one year does not greatly depend on their occurrence in the previous year. Small mammals and many insects with highly mobile populations provide another example. Even for these species, however, permanent quadrats may still outperform temporary quadrats if recruitment most often takes place near parents.

Permanent Sampling Units to Estimate Density

Let us examine two very different situations involving permanent density quadrats. Figure 8.16 compares sample sizes needed to detect different levels of change in density in a clumped population of 4000 plants using permanent and temporary quadrats. All sampling was done with

0.25m × 50m quadrats. In this example, there was no recruitment of new plants; all change between year 1 and year 2 was the result of plant mortality. This created a strong correlation between quadrat counts between the two periods for the low mortality changes. The x-axis shows the percent change in mean plant density (equivalent to percent mortality in this example). The y-axis shows the number of quadrats that needed to be sampled to detect the true population change with false-change and missed-change error rates both set at 0.10. When the change in mean plant density between the first and second sampling periods was less than 50%, permanent quadrats were much more effective than temporary quadrats at tracking the change. For example, for detecting a 5% change, 22 permanent quadrats performed as well as 338 temporary ones!

The advantage of permanent quadrats occurs when counts between two periods correlate with one another. This is true in the situation depicted in Figure 8.16 because no new plants show up in new locations. The opposite extreme, illustrated by Figure 8.17, shows population changes caused by 100% mortality of the original population combined with various levels of recruitment from plants in completely new positions. Permanent quadrats no longer provide any advantage over temporary ones, and the disadvantages of permanent quadrats would lead you to a temporary quadrat design.

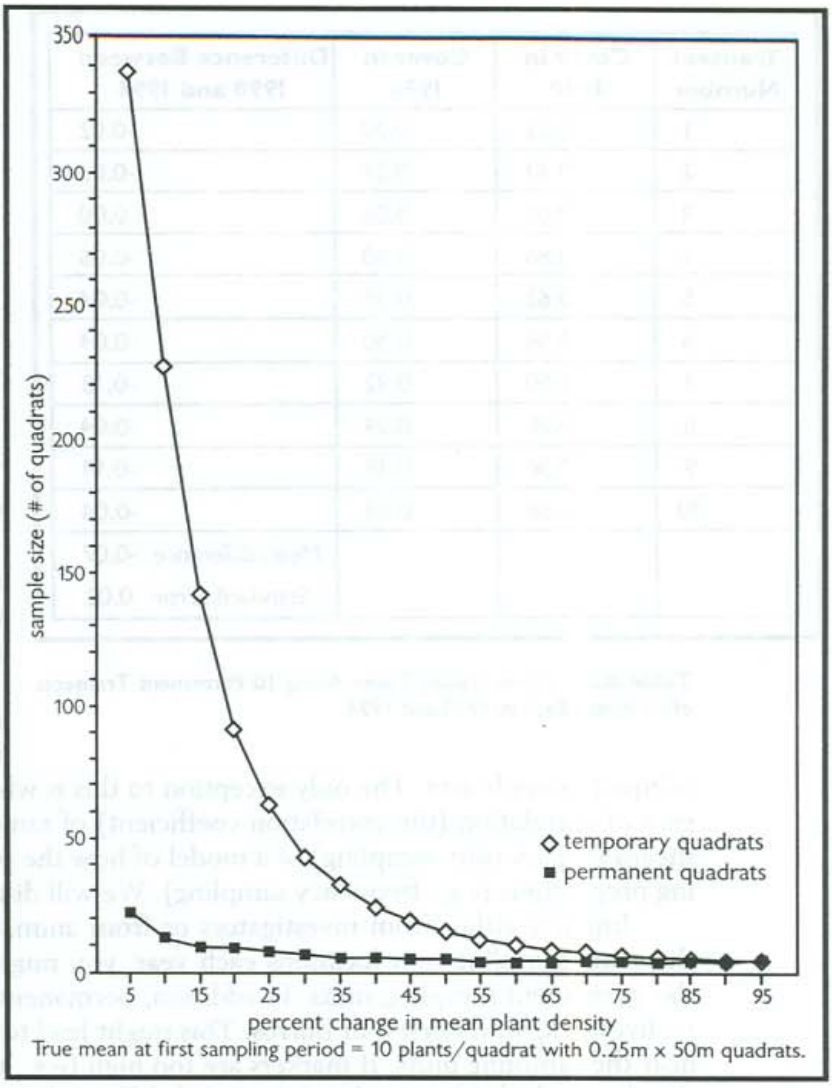


Figure 8.16. Sample sizes needed to detect different degrees of population decline from an artificial clumped population of 4,000 plants using temporary vs. permanent quadrats. All changes are due to mortality of the original population without any recruitment of new plants. Note the much better performance of permanent quadrats in detecting changes below 50%.

Most populations will show a combination of mortality and recruitment, as opposed to the extreme situations shown in Figures 8.16 and 8.17. For most species, permanent quadrats will provide greater precision with the same number of quadrats or equivalent precision at smaller sample sizes, because the locations of new individuals will likely be correlated with the location of old individuals given typical patterns of reproduction. You must balance the magnitude of this increase in precision (or reduction in sample size) against the disadvantages of using permanent sampling units.

Permanent Frequency Quadrats and Points

The discussion so far has centered on the use of paired quadrats for estimating density. This type of sampling is analyzed by means of a paired *t*-test (this will be covered in Chapter 9). The paired *t*-test would also be used to analyze changes in paired quadrats used to estimate cover and to analyze changes in permanent transects such as those used for line intercept sampling or for

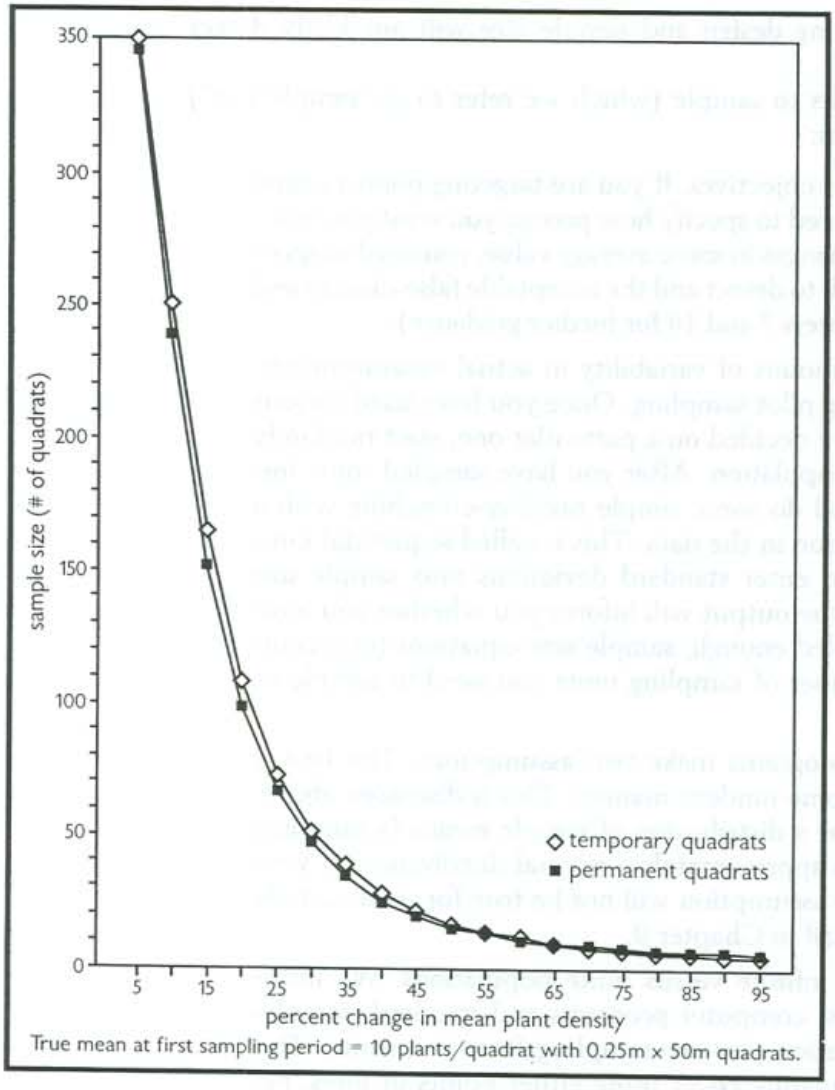


Figure 8.17. Sample sizes needed to detect different degrees of population decline from an artificial clumped population of 4,000 plants using temporary vs. permanent quadrats. All changes result from 100% mortality of the original population with various levels of random recruitment. Temporary and permanent quadrats perform about the same in this situation.

point or quadrat sampling in systematic sampling designs (when the transects, as opposed to the quadrats or points, are treated as the sampling units).

When frequency quadrats or points are treated as the sampling units, a different set of tests is used to determine if a statistically significant change has taken place. The chi-square test is used when these types of sampling units are temporary (i.e., randomly located in each year of measurement), while McNemar's test is used when the quadrats or points are permanently located in the first year of measurement. These tests are discussed in Chapter 9, but it is important here to point out that—just as for permanent designs that use transects or quadrats for estimating density or cover—it is sometimes much more efficient to make use of permanent frequency quadrats or points.

Salzer (unpublished data) concludes that under certain population-change scenarios, permanent frequency quadrats offer large reductions in sample size over those required for temporary quadrats. In the most extreme example, 87 permanent quadrats perform as well as 652 temporary quadrats in detecting a 5.5% decline in frequency (with the false-change and missed-change error rates both set at 0.10). In other situations, little difference exists between permanent quadrat designs and temporary quadrat designs.

The sample-size differences between temporary and permanent frequency designs depend on the particular nature of population changes. For this reason, the determination of whether to use permanent or temporary frequency quadrats must be evaluated on a case-by-case basis, taking into account the life history of the species, the sample size advantages of using the permanent design, and the disadvantages associated with designs using permanent quadrats.

Appendix II contains more information on the use of permanent frequency designs and should help you decide when to use one.

HOW MANY SAMPLING UNITS SHOULD BE SAMPLED?

An adequate sample is vital to the success of any successful monitoring effort. Adequacy relates to the ability of the observer to evaluate whether the management objective has been achieved. It makes little sense, for example, to set a management objective of increasing the density of a

rare plant species by 20% when the monitoring design and sample size will not likely detect changes in density of less than 50%.

Deciding on the number of sampling units to sample (which we refer to as "sample size") should be based on the following considerations:

1. Sample size should be driven by specific objectives. If you are targeting point-in-time estimates (parameter estimation), you need to specify how precise you want your estimates to be. If you are trying to detect changes in some average value, you need to specify the magnitude of the change you wish to detect and the acceptable false-change and missed-change error rates (refer to Chapters 7 and 14 for further guidance).
2. Sample size should be based on the amount of variability in actual measurements. You should assess this variability during pilot sampling. Once you have tried various sampling-unit sizes and shapes and have decided on a particular one, start randomly positioning the sampling units in the population. After you have sampled some initial number of sampling units, stop and do some simple number-crunching with a hand calculator and evaluate the variation in the data. This is called sequential sampling and is discussed below. You can enter standard deviations into sample-size equations or computer programs, and the output will inform you whether you have sampled enough. If you have not sampled enough, sample size equations (or a computer program) will calculate the number of sampling units you need to sample to meet your objective.
3. Sample-size formulas and computer programs make two assumptions. The first is that sampling units are positioned in some random manner. This is discussed above. The formulas and programs also assume a distribution of sample means (a sampling distribution) from your population fits approximately a normal distribution. If your population is highly skewed, this latter assumption will not be true for small sample sizes. We discuss this issue in more detail in Chapter 9.
4. Sample sizes required differ between infinite versus finite populations. We introduced this concept in Chapter 7. Most computer programs and standard, sample-size equations assume that the population you are sampling from is infinite. This will always be the case if you are estimating cover using either points or lines, because these are considered dimensionless. If, however, you are sampling a relatively small area, and you are making density, frequency, cover, or biomass assessments in quadrats, then you should account for the fact that you are sampling from a finite population. This means there is some finite number of quadrats that can be placed in the area to be sampled. The sample-size formulas provided in Appendix II include a correction factor called the Finite Population Correction (FPC). If you are sampling more than 5% of a population, applying the FPC "rewards" you by reducing the necessary sample size. Appendix II describes how to apply the FPC to sample-size determination.[7]
5. Precision increases with sample size but not proportionately. This is illustrated in Figure 8.18 in an example where the statistical benefits of increasing sample size diminish once you reach about $n = 30$. You should seek to increase statistical precision and power not by simply increasing sample size, but by reducing the standard deviation to as small a value as possible through good design.
6. Problems may occur in the use of many published sample-size formulas. Most formulas that are designed to determine sample sizes for "point-in-time" estimates

[7] *Chapter 9 describes how to apply the finite correction factor to results of significance tests. Appendix III describes how to apply the finite correction factor to analysis of confidence intervals.*

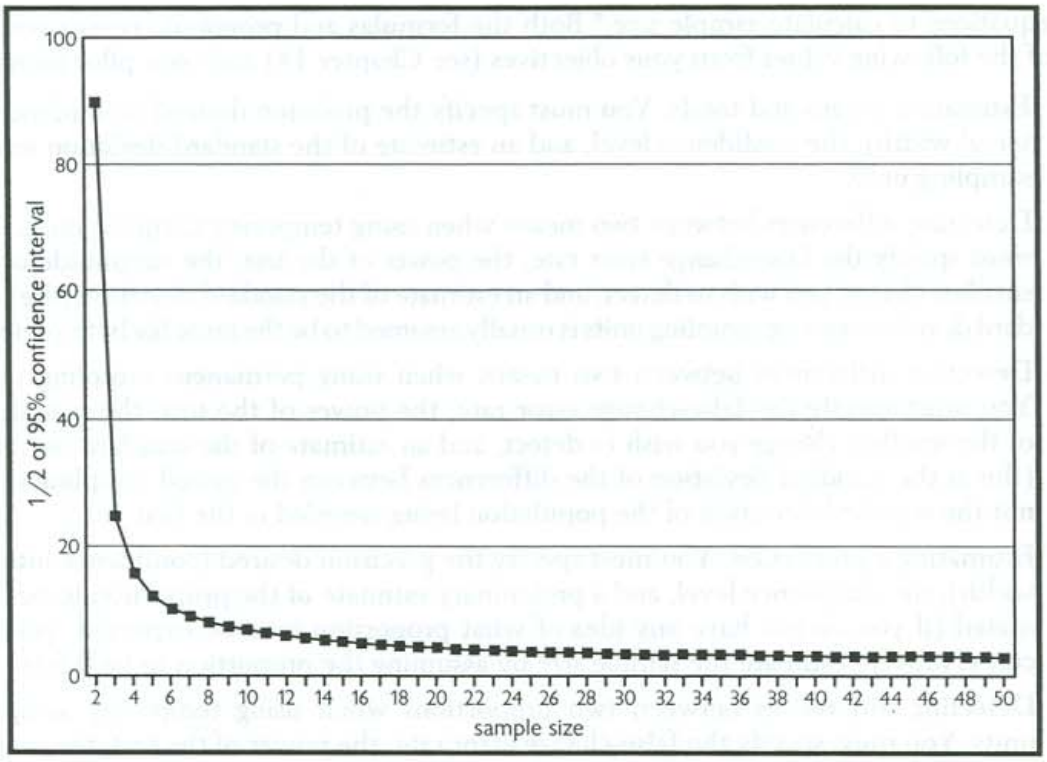


Figure 8.18. Influence of sample size on level of precision. Sample sizes necessary to achieve different levels of precision at a constant standard deviation of 10. Note that there is no effective improvement in precision after about n = 30.

(parameter estimation) with specified levels of precision do not account for the random nature of sample variances. They do not include a "level of assurance" (also known as a tolerance probability) that you will actually achieve the conditions specified in the sample-size equations and obtain a confidence interval of a specified width. Blackwood (1991) provides a layperson's discussion of this topic and reports the results of a simulation that illustrates the concept. Kupper and Hafner (1989) provide a correction table to use with standard, sample-size equations for estimates of single population means or population totals. A modified version of this table and instructions on how to use it are included in Appendix II.

Information Required for Calculating Sample Size

Appendix II gives equations for calculating sample sizes for the following sampling objectives:

1. Estimating means and totals
2. Detecting differences between two means when using temporary sampling units
3. Detecting differences between two means when using permanent sampling units
4. Estimating a proportion
5. Detecting differences between two proportions when using temporary sampling units
6. Detecting differences between two proportions when using permanent sampling units

Equations for calculating sample size for cluster samples, two-stage samples, and stratified random samples are given in Appendix IV. Computer programs are also available that implement

these equations to calculate sample size.[8] Both the formulas and programs require you to insert some of the following values from your objectives (see Chapter 14) and your pilot sampling:

- Estimating means and totals. You must specify the precision desired (confidence interval width), the confidence level, and an estimate of the standard deviation among sampling units.
- Detecting differences between two means when using temporary sampling units. You must specify the false-change error rate, the power of the test, the magnitude of the smallest change you wish to detect, and an estimate of the standard deviation (the standard deviation among sampling units is usually assumed to be the same for both periods).
- Detecting differences between two means when using permanent sampling units. You must specify the false-change error rate, the power of the test, the magnitude of the smallest change you wish to detect, and an estimate of the standard deviation (this is the standard deviation of the differences between the paired sampling units, not the standard deviation of the population being sampled in the first year).
- Estimating a proportion. You must specify the precision desired (confidence interval width), the confidence level, and a preliminary estimate of the proportion to be estimated (if you do not have any idea of what proportion is to be expected, you can conservatively estimate the sample size by assuming the proportion to be 0.50).
- Detecting differences between two proportions when using temporary sampling units. You must specify the false-change error rate, the power of the test, the magnitude of the smallest change you wish to detect, and a preliminary estimate of the proportion in the first year of measurement (using a value of 0.50 will conservatively estimate the sample size).
- Detecting differences between two proportions when using permanent sampling units. You must specify the false-change error rate, the power of the test, the magnitude of the smallest change you wish to detect, and an estimate of the sampling-unit transitions that took place between the 2 years. This last estimate is specific only to this design and is discussed separately in Appendix II.

Your management and sampling objectives already include most of the information required to calculate sample size using either the equations of Appendix II or the computer programs. What is missing is 1) an estimate of the standard deviation, for those situations where you wish to estimate a mean value or detect change between two mean values; and 2) a preliminary estimate of the population proportion, when estimating a proportion or detecting change between two proportions using temporary sampling units. For proportions you have the flexibility of simply entering 0.50 as your preliminary estimate of the population proportion (this provides a conservative estimate of sample size). Alternatively, you can use an estimate derived from pilot sampling. When dealing with mean values, however, you must have an estimate of the standard deviation. This is the subject of the next section.

Sequential Sampling to Obtain a Stable Estimate of the Mean and Standard Deviation

In several places in this chapter we have stressed the need for pilot sampling. The principal purposes of pilot sampling are to assess the efficiency of a particular sampling design and, once a particular design has been chosen, to generate the values needed for calculating the sample size

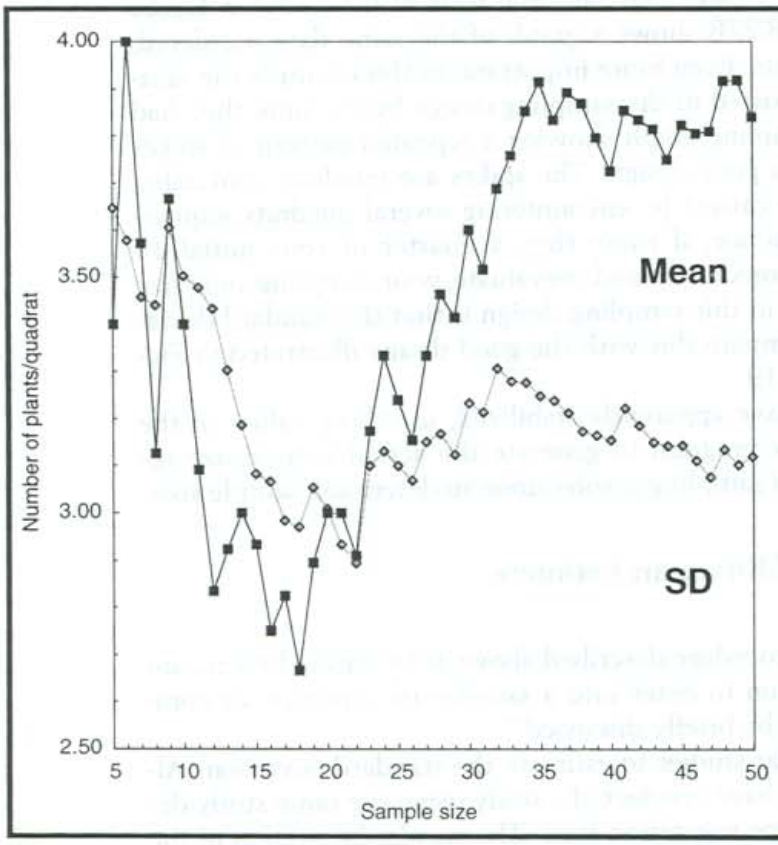
[8] *Surprisingly, many of the general statistical programs, despite their expense, do not include routines for calculating sample size. Thomas and Krebs (1997) reviewed 29 computer programs for calculating sample size; a link to an online copy can be found on our web page. Several freeware or shareware programs are available. Links to these can also be found on our web page (see Preface).*

required to meet the sampling objective. Pilot sampling enables us to obtain stable estimates of the population mean and the population standard deviation and to calculate the **coefficient of variation.** The estimate of the standard deviation derived through pilot sampling is one of the values we use to calculate sample size, whether we use the formulas of Appendix II or a related computer program. Sequential sampling is the process we use to determine whether we have taken a large enough pilot sample to properly evaluate different sampling designs and/or to use the standard deviation from the pilot sample to calculate sample size.

The coefficient of variation (CV) is calculated by dividing the sample standard deviation by the sample mean. The coefficient of variation is useful because, as a measure of variability, it does not depend upon the magnitude and units of measurements of the data. This allows direct comparison of CV's from different studies and of different sampling designs. It also enables us to derive estimates of sample size when we do not have data from pilot studies but do have an idea of the magnitude of CV from similar studies and sites.

We begin by gathering pilot sampling data using some selected sample size. The selection of this initial sample size will depend upon the relative amount of variation in the data—if many of the sampling units yield numbers similar to one another, then you may want to perform the first sequential sampling procedure after $n = 8$ or 10. If you see high variation among the sampling units, then you may want to start with a larger number (e.g., $n \geq 15$) or, perhaps preferably, consider altering the size and/or shape of your sampling unit before doing the first iteration of the sequential sampling procedure.

Calculate the mean and standard deviation for the first two sampling units, calculate it again after putting in the next sampling unit, and then repeat this procedure for all of the sampling units sampled so far. This will generate a running mean and standard deviation. Look at the four columns of numbers on the right of Figure 8.19 for an example of how to carry out this proce-



Running means and standard deviations

n	Plants	Mean	SD	n	Plants	Mean	SD
1	1	1.00		26	1	3.15	3.07
2	2	1.50	0.71	27	8	3.33	3.15
3	0	1.00	1.00	28	7	3.46	3.17
4	9	3.00	4.08	29	2	3.41	3.12
5	5	3.40	3.65	30	9	3.60	3.23
6	7	4.00	3.58	31	1	3.52	3.21
7	1	3.57	3.46	32	9	3.69	3.31
8	0	3.13	3.44	33	6	3.76	3.28
9	8	3.67	3.61	34	7	3.85	3.28
10	1	3.40	3.50	35	6	3.91	3.25
11	0	3.09	3.48	36	1	3.83	3.24
12	0	2.83	3.43	37	6	3.89	3.21
13	4	2.92	3.30	38	3	3.87	3.17
14	4	3.00	3.19	39	1	3.79	3.16
15	2	2.93	3.08	40	1	3.73	3.15
16	0	2.75	3.07	41	9	3.85	3.22
17	4	2.82	2.98	42	3	3.83	3.18
18	0	2.67	2.97	43	3	3.81	3.15
19	7	2.89	3.05	44	1	3.75	3.14
20	5	3.00	3.01	45	7	3.82	3.14
21	3	3.00	2.93	46	3	3.80	3.11
22	1	2.91	2.89	47	4	3.81	3.08
23	9	3.17	3.10	48	9	3.92	3.13
24	7	3.33	3.13	49	4	3.92	3.10
25	1	3.24	3.10	50	0	3.84	3.12

Figure 8.19. A sequential sampling graph. Running means and standard deviations are plotted for increasing sample sizes. Note how the curves smooth out after n = 35.

dure. Most hand calculators enable you to add additional values after you have calculated the mean and standard deviation, so you do not have to input the previous values again.

Use graph paper (or preferably a computer spreadsheet program) to plot the mean and standard deviation against sample size (Fig. 8.19). We suggest starting your graph at $n = 3$ or $n = 5$ for reasons that will become clear later. You are looking for a smoothing of the graph, suggesting the mean and standard deviation have stabilized.

A laptop or field computer is extremely valuable for creating sequential sampling graphs during a pilot sampling program. Spreadsheet programs allow you to enter your data in a form that can later be analyzed (saving time on later data entry) and at the same time create a sequential sampling graph of the running mean and standard deviation. You can also reorder the data (as though you had measured the sampling units in a different order) and replot the sequential sampling graph.

Now, let us apply these concepts. Examine Figure 8.20. The graph shows two sampling sequences of the same population using the same sampling units. The difference between them is that, simply by chance, in the first sequence several plots with large numbers of plants were the first to be sampled, while in the second sequence the first plots to be sampled had only a few plants (or none). Where would you stop sampling in either of these sequences (consider the curve "smoothed")? One strategy would be to reorder the sampling units and evaluate alternative sequential sampling graphs. A better strategy would be to re-evaluate the sampling design. Look again at Figure 7.2 in Chapter 7. Do you think the 0.4m × 10m quadrat is an efficient sampling-unit design? If, after sampling 20% to 30% of the possible sampling units, your sequential sampling graph has not stabilized, you should definitely reconsider your design. Figure 8.21 shows a good sampling design with the curve smoothing at about $n = 12$. The samplers could have saved a substantial amount of effort by stopping long before they did.

Figure 8.22 illustrates the problems that may arise from plotting your graph beginning with the first data point. Here, a large initial value of six plants and the scale of the y-axis in Figure 8.22A give an illusion of smoothing. Figure 8.22B shows a graph of the same data reordered, with the first quadrat containing only two plants. Even more important, in this example the sampler should have recognized that a problem existed in the sampling design by the time they had sampled $n = 20$ (or earlier). A sequential sampling graph showing a repeated pattern of spikes followed by gradual declines is indicative of a poor design. The spikes are quadrats containing many of the species. The gradual declines are caused by encountering several quadrats sequentially with zero observations in them. In practice, if more than a quarter of your initial 10 quadrats contain none of the species, stop immediately and reevaluate your sampling-unit size and shape. Another hint that a problem exists in this sampling design is that the standard deviation is consistently greater than the mean. Compare this with the good design illustrated in Figure 8.20 and the acceptable design of Figure 8.19.

Once the mean and standard deviation have apparently stabilized, use those values in the appropriate sample-size equation or computer program to generate the actual sample size required. We do not recommend using sequential sampling graphs alone to determine sample size.

Alternatives to Sequential Sampling to Obtain an Estimate of the Standard Deviation

Pilot sampling, using the sequential sampling procedure described above, is by far the best means of deriving an estimate of the standard deviation to enter into a sample-size equation or computer program. Two less effective methods will be briefly discussed.

The first method is to use data from similar studies to estimate the standard deviation. Although not as reliable as a pilot study, you may have conducted a study using the same study design and measuring the same attribute in the same vegetation type. The standard deviation of the sample from this study can be used as an estimate of the standard deviation of the population that is the focus of the current study.

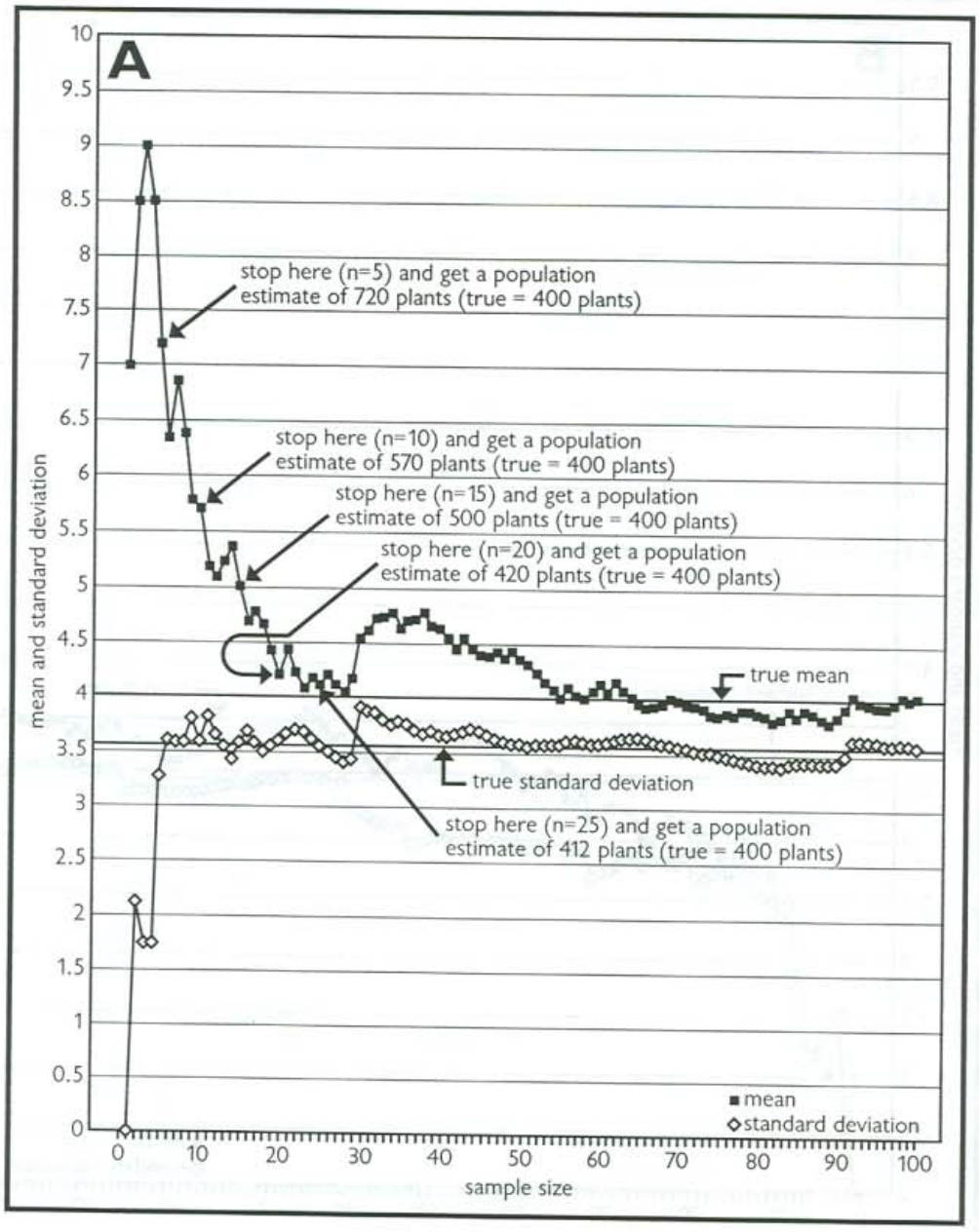


Figure 8.20. Sequential sampling graphs of the 20m × 20m "400 plant population" introduced in Chapter 7. The population was sampled using a 0.4m × 10m quadrat. The entire population consists of 100 quadrats. Notice how far estimates are from the true mean if they are made before the curves smoothing out. In Part A many of the quadrats sampled at the beginning had large values. Note how we would have overestimated the population if we had stopped too soon.

The second method relies on professional judgment. As pointed out by Krebs (1998), an experienced person may have some knowledge of the amount of variability in a particular attribute. Using this information you can determine a range of measurements to be expected (maximum value – minimum value) and can use this range to estimate the standard deviation of a measure. Table 8.4, adapted from the table in Dixon and Massey (1983), gives the appropriate conversion factor to be multiplied by the range value to come up with an estimate of the population standard deviation.

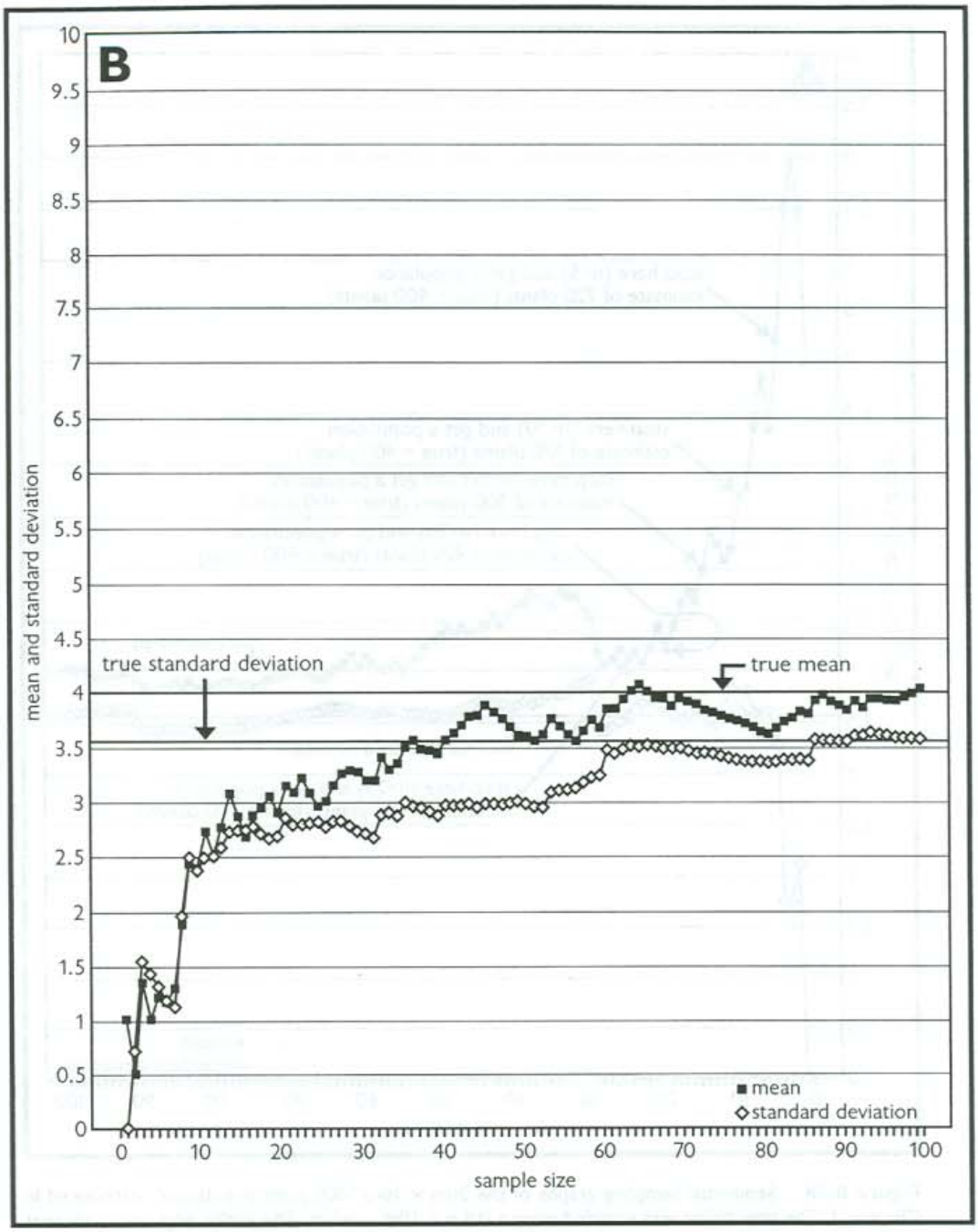


Figure 8.20B. This figure is the same sample as Part A, but with the data randomly reordered. If we'd used the initial values shown in this graph (prior to the curves leveling off), we would have seriously underestimated the true mean value, as opposed to overestimating as in Part A.

To illustrate how to use this table, let us assume we know from working with a particular species that in a sample of size 30 we could expect a range of 0 individuals per quadrat to 100 individuals per quadrat (this process assumes a normal distribution so we should not have too many quadrats with zeros in them). The range in this case is 100 – 0 = 100 individuals. The conversion factor for a sample of size 30 is 0.245. Our estimate of the population standard deviation is, therefore, 100 individuals × 0.245 or 24.5 individuals per quadrat.

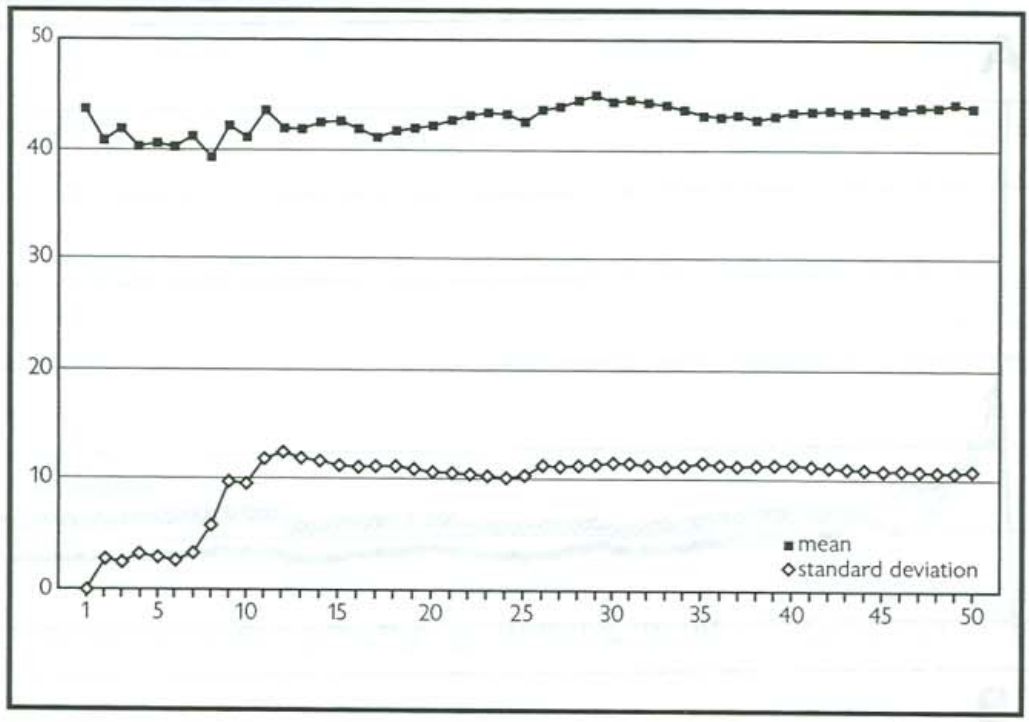


Figure 8.21. Sequential sampling graph of vegetation height measurements. Note how the graphs have flattened out long before the sampling ended.

Although this method can be used, it should be emphasized again that data from a pilot study are more reliable and are preferable to this method.

Estimating the Standard Deviation When Using Permanent Sampling Units

Estimating the standard deviation for a design that uses permanent sampling units is difficult because it is the standard deviation of the difference between the sampling units between the two years that must be entered into the sample size equation or computer program, and this is a value that you will not have until you have collected data in the second year. Thus, your pilot study must span 2 years before you can accurately estimate the sample size required to meet your sampling objective. You would like, however, to make a reasonable estimate from the first year's data of the standard deviation of the difference. This will give you a good chance of having used a large enough sample size the first year, with the result that you will not have to add more sampling units the second year and will be able to use the first year's data in your analysis. Following are some methods you can use for this purpose.

You can estimate the standard deviation using the alternative methods discussed in the section above. Remember, however, that it is the standard deviation of the difference that must be estimated, so if you use data from previous studies, they must be studies that used permanent sampling units. If you use the expected range to estimate the standard deviation, it must be the range of the differences, not the range of the data for any one year.

There is another way you can calculate the necessary sample size by having only the first year's pilot data. This method requires that you have some knowledge of the degree of correlation (correlation coefficient) expected between the permanent sampling units between years. Appendix II provides a formula by which you can estimate the standard deviation of the differ-

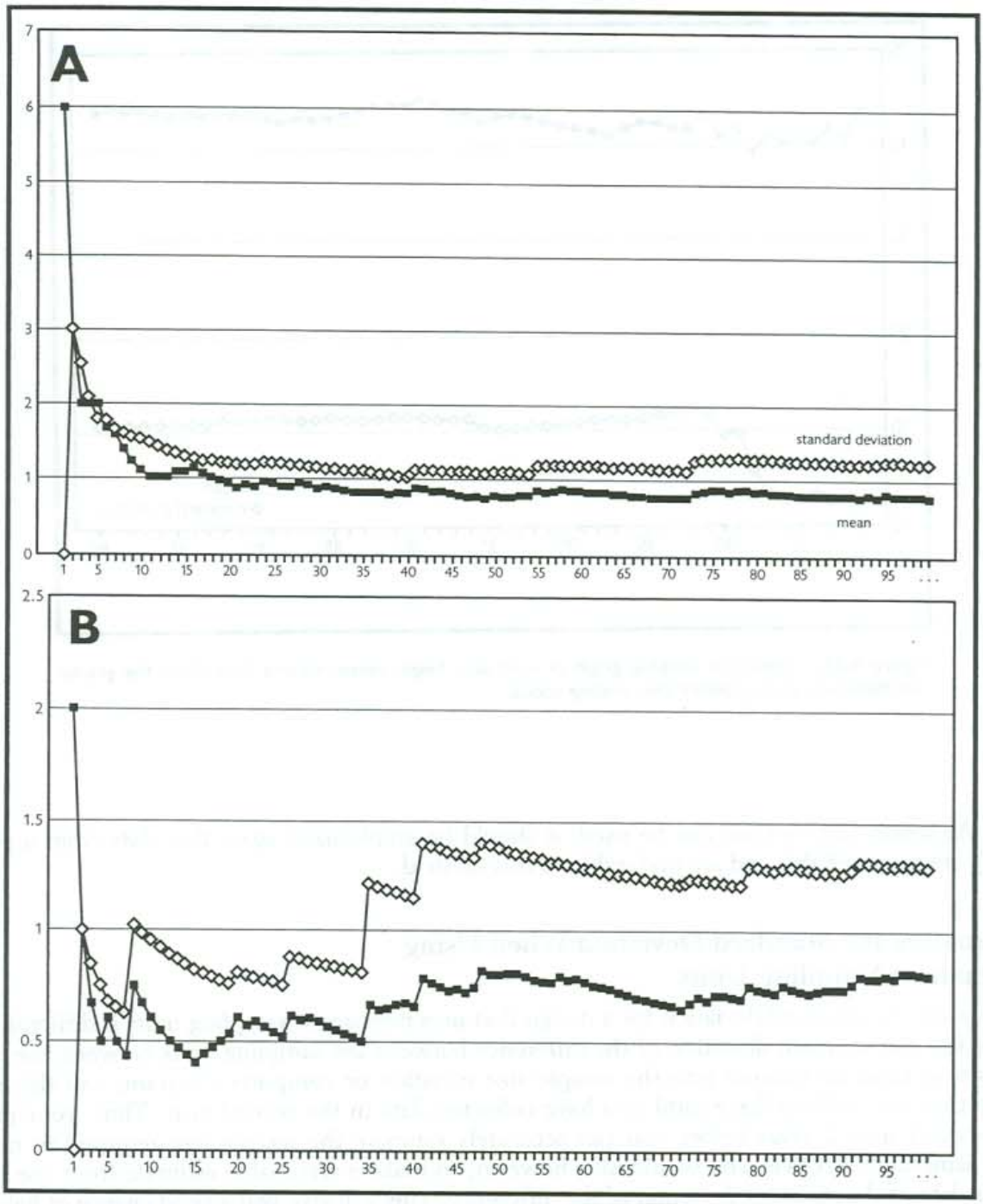


Figure 8.22. Sequential sampling graphs for *Astragalus applegatei* at the Euwana Flat Preserve. Part A shows what can happen when the y-axis is set at too large a range, because of initial large values. This can make it appear that the running mean and standard deviation has smoothed out when in fact they haven't. Part B illustrates the real situation: neither statistic has smoothed out even by n = 100. This is a poor sampling design. See text for further elaboration.

Sample Size	Conversion Factor	Sample Size	Conversion Factor
2	0.886	19	0.271
3	0.591	20	0.268
4	0.486	25	0.254
5	0.430	30	0.245
6	0.395	40	0.231
7	0.370	50	0.222
8	0.351	60	0.216
9	0.337	70	0.210
10	0.325	80	0.206
11	0.315	90	0.202
12	0.307	100	0.199
13	0.300	150	0.189
14	0.294	200	0.182
15	0.288	300	0.174
16	0.283	500	0.165
17	0.279	1000	0.154
18	0.275		

Table 8.4. Conversion Factors Used to Estimate the Population Standard Deviation. To estimate the standard deviation of a variable from knowledge of the range for samples of various sizes, multiply the observed range (maximum – minimum value) by the table values to obtain an unbiased estimate of the standard deviation. This procedure assumes a normal distribution. From Dixon and Massey (1983) and reproduced in Krebs (1998).

ence between years by using the standard deviation of the first year's sample and the correlation coefficient. This is something you might have from similar studies on the same species (although in that case you would probably already have an estimate of the standard deviation of the difference between years that you could use). Based on your knowledge of the life history of the species you are dealing with, you might make an initial estimate of correlation. For example, if you are monitoring a long-lived perennial and do not anticipate a lot of seedling recruitment (or if you expect seedling recruitment to be very close to parent plants), you might estimate that the correlation coefficient between years is relatively high, say about 0.80 or 0.90. You then plug this coefficient into the formula, along with your estimate of the standard deviation of the first year's data.

Whichever method you use to estimate the standard deviation of the difference, once you have collected the second year's data, you will still need to enter the actual observed standard deviation of the difference into an equation or a computer program to calculate actual sample size. You can then modify your initial estimate of sample size accordingly.

MANAGEMENT IMPLICATIONS

Good sampling design can dramatically increase the precision of the estimates of population characteristics while reducing field costs. While good design may be time-consuming at the planning stage of a monitoring study, the investment pays well throughout the life of the monitoring and in the application of data to management decisions. Six design features must be addressed when planning a monitoring study using sampling:

1. What is the population of interest?
2. What is an appropriate sampling unit?
3. What is an appropriate sampling-unit size and shape?
4. How should sampling units be positioned?
5. Should sampling units be permanent or temporary?
6. How many sampling units should be sampled?